PHARMACEUTICAL COMPOSITION COMPRISING A BENZODIAZEPINE DERIVATIVE AND A INHIBITOR OF THE RSV FUSION PROTEIN

The present invention relates to a series of anti-viral benzodiazepine derivatives. In particular, it relates to a series of benzodiazepine derivatives which interact with an inhibitor of the RSV fusion protein to provide an additive or synergistic therapeutic effect in treating or preventing an RSV infection.

Respiratory Syncytial Virus (RSV) is a major cause of respiratory illness in patients of all ages. In adults, it tends to cause mild cold symptoms. In school-aged children, it can cause a cold and bronchial cough. In infants and toddlers it can cause bronchiolitis (inflammation of the smaller airways of the lungs) or pneumonia. It has also been found to be a frequent cause of middle ear infections (otitis media) in pre-school children. RSV infection in the first year of life has been implicated in the development of asthma during childhood.

Current anti-RSV therapy involves the use of a monoclonal antibody to RSV, called palivizumab. Such use of palivizumab is a prophylactic, rather than therapeutic, treatment of RSV. However, although this antibody is often effective, it is expensive. Indeed, its expense means that it is unavailable for many people in need of anti-RSV therapy. There is therefore an urgent need for effective alternatives to existing anti-RSV therapy.

Small compounds which inhibit RSV replication by inhibiting the fusion (F) protein of RSV block the entry of the virus into the host cell and the exit from the host cell via syncytia formation. While these compounds have been shown to have high potency, RSV rapidly develops resistance to these compounds through mutations in the F protein (Morton, C.J. et al, 2003.Virology 311, 275-288).

PCT/GB03/04050 filed on 20 September 2003 discloses a series of benzodiazepine derivatives which inhibit RSV replication. Serial passaging experiments have indicated that resistance to these inhibitors is slow to develop and sequencing of resistant mutants did not reveal any significant changes in the F protein. It can therefore be assumed that these benzodiazepines have a common and novel mode of action, which does not involve inhibition of the F-protein.

It has now surprisingly been shown that a combination of (a) an RSV fusion protein inhibitor and (b) an anti-RSV benzodiazepine is highly active against RSV. Components (a)

and (b) are found to have at least an additive effect. Further, it is also a finding of the invention that the two components interact synergistically, to provide a combined effect that is greater than the sum of the effects of the individual components.

The present invention therefore provides, in a first embodiment, a pharmaceutical composition which comprises a pharmaceutically acceptable carrier or diluent and

(a) an inhibitor of the RSV fusion protein; and:

(b) a benzodiazepine derivative capable of inhibiting RSV replication.

It is a finding of the present invention that components (a) and (b) have at least an additive effect. The concepts of synergism and additivity are, of course, well known in the field of pharmacology. It is thus well established that a therapeutically useful additive combination is one in which the effect of the combination is greater than the larger of the effects produced by each of the components at the same concentrations as in the mixture. Thus, in the present case, a given formulation containing x wt% of component (a) and y wt% of component (b) has an activity which is at least as great as the activity of a formulation containing, as sole active ingredient, either x wt% component (a) or y wt% component (b).

In such additive combinations, the active ingredients are typically operating via different physiological pathways. In the present case, for example, component (a) and component (b) are believed to be inhibiting separate RSV proteins. An additive combination is therapeutically useful because it can achieve a therapeutically useful effect using lower concentrations of each active component. This enables the side-effects of the medication to be minimised. Thus, the additive combination can be formulated so that each active ingredient is present at a concentration which is subclinical in cells other than the target disease cells. The additive combination is nevertheless therapeutically effective in target cells which respond to both ingredients.

As regards component (a), an inhibitor of the RSV fusion protein can be identified by an assay comprising:

(a) labelling RSV with octadecyl rhodamine dye (R18);

(b) pre-incubating the labelled virus with Hep-2 cells seeded in a 6-well plate at 1 hour for 4°C;

(c) removing unattached virus;

(d) adding the candidate fusion protein inhibitor;

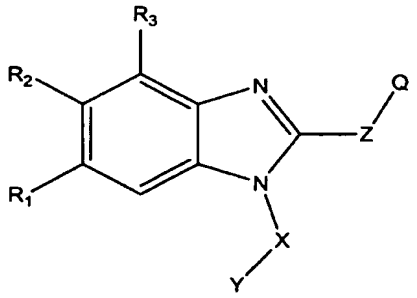
(e) incubating the 6-well plates at 37°C for 1 hour; and

(f) determining any increase in fluorescence, typically using a fluorescence microscope.

In the above assay, any increase in fluorescence signifies a fusion event. Thus, if no increase in fluorescence is detected, 100% inhibition is achieved. If the increase in fluorescence is equal to that observed with a corresponding assay in which a control of growth medium and solvent (e.g., growth medium with 10% fetal bovine serum and DMSO) is used in step (d) in place of the candidate fusion protein inhibitor, 0% inhibition is achieved. Accordingly the % inhibition achieved with the candidate fusion protein inhibitor can be determined by quantitative assessment of the fluorescence in step (f).

As used herein, component (a) is typically a compound which achieves at least 10%, more typically at least 30%, preferably at least 50% and most preferably at least 75%, inhibition of the RSV fusion protein as determined by the above assay.

Typically, component (a) is a compound of formula (I), or a pharmaceutically acceptable salt thereof,

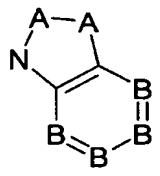


(I)

wherein:

- X is a direct link or C_{1-6} alkyl; said C_{1-6} alkyl being optionally substituted with halogen, oxo, cyano, hydroxyl, $OCOR_4$ or $S(O)n\text{-}C_{1-6}$ alkyl;
- Y is R_4, NR_4R_5, $NCOR_4$, $=N\text{-}OR_4$, $-CONHR_4$, $COOR_4$, $-OR_4$, aryl, heteroaryl, cyclyl or heterocyclyl, where R_4 and R_5 are H or C_{1-6} alkyl;
- Z is CR_6R_7, where R_6 and R_7 are independently H, or straight, branched or cyclic C_{1-6} alkyl;
- n is 1-2;

- R_1 is $CONR_4R_5$, CO_2R_4 or C_{1-6} alkyl, said C_{1-6} alkyl can be optionally substituted with OR_4 or NR_8R_9;
- R_8 and R_9 are each independently H, C_{1-6} alkyl, SO_2R_5, CO_2R_4 or COR_4;
- R_2 is selected from the group consisting of NH_2, $CONR_6R_7$, heteroaryl, C_{2-6} alkenyl, CO_2R_4, $N{=}CPh_2$, $C(=NH)NH_2$ and C_{1-6} alkyl; said alkyl optionally substituted with a member selected from the group consisting of halogen, CN, $NR_{10}R_{11}$, OSO2R_4 and OR_4;
- R_{10} and R_{11} are each independently selected from the group consisting of H, C_{1-6} alkyl, C_{3-6} cycloalkyl, CO_2R_4, COR_4 and SO_2R_4;
- R_3 is selected from the group consisting of (1) CO_2R_9; (2) C_{1-6} alkyl optionally substituted with CN, OR_4 or NR_6R_7; and (3) C_{2-6} alkenyl substituted with CN;
- Q is a member selected from the group consisting of



A is C or N, optionally substituted with H, halogen, C_{1-6} alkyl, C_{2-6} alkenyl, cyano-C_{1-6} alkyl, CO_2R_4, aryl, benzoaminocarbonyl, hydroxybenzyl, $SO_2NR_4R_5$ or C_{3-6} cycloalkyl. Where A is carbon, it may also be optionally substituted by O or S via a double bond;

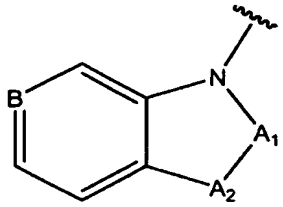
B is C or N; where B is C it may be optionally substituted by H, C_{1-6} alkyl, NO_2, CN, halogen, COR_4, $COOR_4$, $CONHR_4C(=NH)NH_2$ or $C(=NOH)NH_2$.

Typically, at least two of R_1, R_2 and R_3 are hydrogen, and the other is hydrogen or -C(NH)-NH_2. Preferably, all of R_1, R_2 and R_3 are hydrogen.

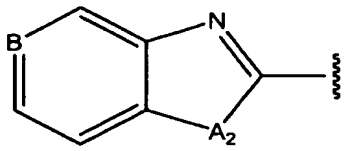
Typically, either -X-Y is H, or X is a C_1-C_6 alkylene group which is unsubstituted or substituted by a hydroxy group and Y is H, OH, CN, -$NR'R''$, -COR', -SO_2R' or phenyl, wherein R' and R'' are the same or different and represent a C_1-C_4 alkyl group.

Typically, Z is -CH_2-.

Typically, Q is a moiety

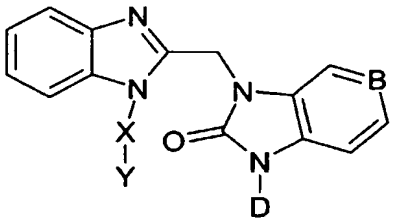


or



wherein B is -CH- or -N-, A_1 is -C(O)- or -NH- and A_2 is $-CH_2-$, $-CHR'-$ or $-NR''-$, wherein R' is a halogen atom and R'' represents a hydrogen atom or a C_1-C_4 alkyl, C_2-C_4 alkenyl, C_3-C_6 cycloalkyl, $-SO_2-(C_1-C_6$ alkyl), $-SO_2-N(C_1-C_6$ alkyl$)_2$ or $-(CO-NH)_a-(C_1-C_4$ alkyl)-phenyl group, wherein a is 0 or 1, which group is unsubstituted or is substituted with a hydroxy or cyano substituent.

Particularly preferred compounds of the invention are compounds of formula (Ia) and pharmaceutically acceptable salts thereof

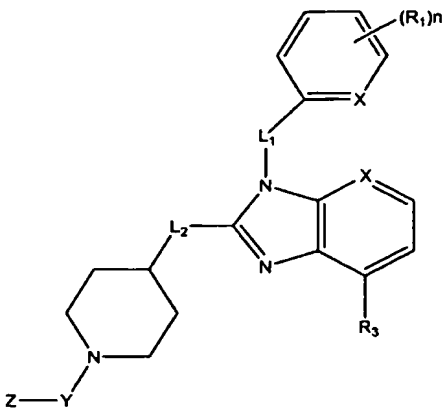


(Ia)

wherein

- B, X and Y are as described in formula (I) above
- D is cyclopropyl, ethyl, 4-cyanobutyl, isopropenyl, methylsulfonyl, dimethylsulfamoyl, benzylaminocarbamoyl or para-hydroxybenzyl

Component (a) can also be a compound of formula (II), or a pharmaceutically acceptable salt thereof,



(II)

wherein:

- L_1 is $-CH_2-$ or $-CHR_2-CO-$
- each X is the same or different and CH or N;
- each R_1 is the same or different and is C_{1-6} alkyl, halogen, hydroxy, phenyl or $(CH_2)_m=NH_2$;
- n is 1 or 2;
- R_2 is C_{1-6} alkoxy or C_{1-6}alkoxy-phenyl;
- R_3 is C_{1-6}alkyl;
- L_2 is $-CH_2-$ or -NH-;
- Y is C_{1-6} alkyl or C_{1-6} alkenyl;
- Z is H, $N(R_4)_2$, $-C(=O)-R_5$, $-C(=CH_2)-R_5$, $-CH(OH)-R_5$, $-CH(CH3)-R_5$, $-CH(OCH3)-R_5$;
- each R_4 is the same or different and is H, C_{1-6} alkyl;
- R_5 is C_{1-6} alkyl-carbonyl, amino, hydroxyl, aryl, heteroaryl, carbocyclyl, heterocyclyl;

and

- m=1-6

For the avoidance of doubt, when L_1 is $-CHR_2-CO-$, the carbonyl group is attached to the phenyl or pyridyl moiety.

Typically, L_1 is $-CH_2-$.

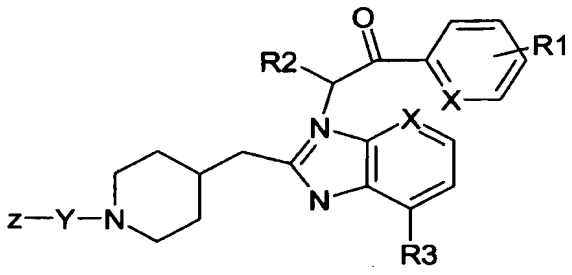
Typically, L_2 is -NH-.

Typically, R_1 is methyl or hydroxy. Typically, n is 2. Typically, each R_1 is different.

Typically, Y is C_1-C_4 alkyl.

Typically, Z is -NH_2.

Other preferred compounds of formula (II) are compounds of formula

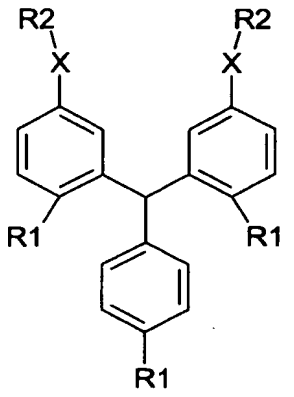


(II)

wherein:

- X is C or N;
- R_1 is C_{1-6} alkyl, halogen, phenyl or $(CH_2)_m$=NH_2;
- R_2 is C_{1-6} alkoxy or C_{1-6}alkoxy-phenyl;
- R_3 is C_{1-6}alkyl;
- Y is C_{1-6} alkyl or C_{1-6} alkenyl;
- Z is H, NR_4, -C(=O)-R_5, -C(=CH_2)-R_5, -CH(OH)-R_5, -CH(CH3)-R_5, -CH(OCH3)-R_5;
- R_4 is H, C_{1-6} alkyl.
- R_5 is C_{1-6} alkyl-carbonyl, amino, hydroxyl, aryl, heteroaryl, carbocyclyl, heterocyclyl
- m=1-6

Component (a) can also be a compound of formula (III), or a pharmaceutically acceptable salt thereof,

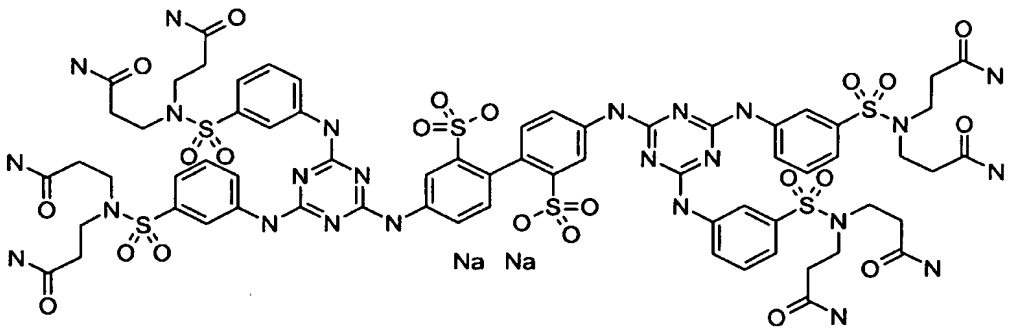


(III)

wherein

- X is –N=C- or -CH=CH-;
- R_1 is H, hydroxyl, alkyl, halogen, nitro or alkoxy; said alkoxy being optionally monosubstituted with carboxy, amino, monoalkylamino, dialkylamino or acetoamino;
- R_2 is pyrazolyl, triazolyl or tetrazolyl and optionally substituted by amino or alkyl.

Component (a) can also be a compound of formula (IV), or a pharmaceutically acceptable salt thereof.



The compound of formula (IV) is 4,4'-Bis-(4,6-bis-{3-[bis-(2-carbamoyl-ethyl)-sulfamoyl]-phenylamino}-[1,3,5]triazin-2-ylamino)-biphenyl-2,2'-disulfonic acid.

Preferably, component (a) is:

1-Cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one

{2-[2-(1,2-Dihydro-benzotriazol-1-ylmethyl)-benzoimidazol-1-yl]]ethyl}-diethyl-amine

{2-[2-(3-Iodo-2,3-dihydro-indazol-1-ylmethyl)-benzimidazol-1-yl]-ethyl}-dimethyl-amine

1-Isopropenyl-3-[1-(3-methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one

1-(4-Hydroxy-benzyl)-3-[1-(3-methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one

1-Isopropenyl-3-[1-(3-oxo-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one

1-Ethyl-3-[1-(2-hydroxy-2-phenyl-ethyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one

1-Ethyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one

7-[2-(3-Isopropenyl-2-oxo-2,3-dihydrobenzoimidazol-1-ylmethyl)-benzoimidazol-1-yl]-heptanenitril

5-{3-[1-(3-Methanesulfonyl-propyl)-1H-benzoimidazol-2-ylmethyl]-2-oxo-2,3-dihydro-benzoimidazol-1-yl}-pentanenitrile

3-[1-(3-Methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-2-oxo-2,3-dihydro-benzoimidazol-1-carboxylic acid benzylamide

1-Methanesulfonyl-3-[1-(3-methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one

3-[1-(3-Methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-2-oxo-2,3-dihydro-benzoimidazol-1-sulfonic acid dimethylamide

1-Isopropenyl-3-(1-propyl-1H-benzoimidazol-2-ylmethyl)-1,3-dihydro-imidazo[4,5-c]pyridine-2-one

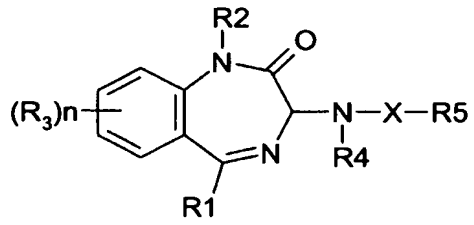
Bis(5-amidino-2-benzimidazolyl)-methane

2-{2-[1-[1-(2-Amino-ethyl)-piperidin-4-ylamino]-4-methyl-benzoimidazol-1-ylmethyl}-6-methyl-pyridin-3-ol

or a pharmaceutically acceptable salt thereof.

In a further embodiment, the composition contains an RSV fusion inhibitor, as described above, and a benzodiazepine identifiable as having anti-RSV activity by the method of Example 8.

Typically, component (b) is a compound of formula (V), or a pharmaceutically acceptable salt thereof,



(V)

wherein:

- R^1 represents C_{1-6} alkyl, aryl or heteroaryl;
- R^2 represents hydrogen or C_{1-6} alkyl;
- each R^3 is the same or different and represents halogen, hydroxy, C_{1-6} alkyl, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, amino, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, nitro, cyano, $-CO_2R'$, $-CONR'R''$, $-NH-CO-R'$, $-S(O)R'$, $-S(O)_2R'$, $-NH-S(O)_2R'$, $-S(O)NR'R''$ or $-S(O)_2NR'R''$, wherein each R' and R'' is the same or different and represents hydrogen or C_{1-6} alkyl;
- n is from 0 to 3;
- R^4 represents hydrogen or C_{1-6} alkyl;
- X represents -CO-, $-CO-NR'-$, -S(O)- or $-S(O)_2-$, wherein R' is hydrogen or a C_1-C_6 alkyl group; and
- R^5 represents an aryl, heteroaryl or heterocyclyl group which is substituted by a C_1-C_6 hydroxyalkyl group or a $-(C_1$-C_4 alkyl)-X_1-(C_1-C_4 alkyl)-X_2-(C_1-C_4 alkyl) group, wherein X_1 represents -O-, -S- or $-NR'-$, wherein R' represents H or a C_1-C_4 alkyl group, and X_2 represents -CO-, -SO- or $-SO_2-$, or R_5 represents $-A_1-Y-A_2$, wherein:
- A_1 is an aryl, heteroaryl, carbocyclyl or heterocyclyl group;
- Y represents a direct bond or a C_1-C_4 alkylene, $-SO_2-$, -CO-, -O-, -S- or $-NR'-$ moiety, wherein R' is a C_1-C_6 alkyl group; and

- A_2 is an aryl, heteroaryl, carbocyclyl or heterocyclyl group.

As used herein, a C_{1-6} alkyl group or moiety is a linear or branched alkyl group or moiety containing from 1 to 6 carbon atoms, such as a C_{1-4} alkyl group or moiety. Examples of C_{1-4} alkyl groups and moieties include methyl, ethyl, *n*-propyl, *i*-propyl, *n*-butyl, *i*-butyl and *t*-butyl. For the avoidance of doubt, where two alkyl moieties are present in a group, the alkyl moieties may be the same or different.

As used herein, a hydroxyalkyl group is typically a said alkyl group that is substituted by one or more hydroxy groups. Typically, it is substituted by one, two or three hydroxy groups. Preferably, it is substituted by a single hydroxy group. A preferred hydroxyalkyl group is $-CH_2-OH$.

As used herein, an acyl group is a C_{2-7} acyl group, for example a group -CO-R, wherein R is a said C_{1-6} alkyl group.

As used herein, an aryl group is typically a C_{6-10} aryl group such as phenyl or naphthyl. Phenyl is preferred. An aryl group may be unsubstituted or substituted at any position. Typically, it carries 0, 1, 2 or 3 substituents.

Suitable substituents on an aryl group include halogen, C_{1-6} alkyl, C_{2-7} acyl, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, nitro, cyano, carbamoyl, mono(C_{1-6} alkyl)carbamoyl, di(C_{1-6} alkyl)carbamoyl, amino, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, $-CO_2R'$, $-CONR'R''$, $-S(O)R'$, $-S(O)_2R'$, $-S(O)NR'R''$,$-S(O)_2NR'R''$ $-NH-S(O)_2R'$ or $-NH-CO-R'$, wherein each R' and R'' is the same or different and represents hydrogen or C_{1-6} alkyl.

Preferred substituents on an aryl group include halogen, C_{1-6} alkyl, C_{2-7} acyl, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, amino, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, nitro, cyano, $-CO_2R'$, $-S(O)R'$, $-S(O)_2R'$ and $-S(O)_2NR'R''$, wherein each R' and R'' is the same or different and represents hydrogen or C_{1-4} alkyl.

Particularly preferred substituents include fluorine, chlorine, bromine, iodine, cyano, C_{1-4} alkyl, C_{2-4} acyl, hydroxy, C_{1-4} alkoxy, C_{1-4} alkylthio, C_{1-4} haloalkyl, C_{1-4} haloalkoxy, amino, mono(C_{1-4} alkyl)amino, di(C_{1-4} alkyl)amino, nitro, $-CO_2R'$, $-S(O)_2R'$ and $-S(O)_2NH_2$, wherein R' represents C_{1-2} alkyl. Most preferred substituents are chlorine, fluorine, cyano, C_1-C_4 alkyl and C_1-C_4 haloalkyl substituents.

As used herein, references to an aryl group include fused ring systems in which an aryl group is fused to a monocyclic carbocyclyl, heterocyclyl or heteroaryl group or to a fused group which is a monocyclic carbocyclyl, heterocyclyl or heteroaryl group which is fused to a phenyl ring. Typically, said fused ring systems are systems in which an aryl group is fused to a monocyclic carbocyclyl, heterocyclyl or heteroaryl group.

Preferred such fused ring systems are those wherein an aryl group is fused to a monocyclic heterocyclyl or heteroaryl group or to a monocyclic carbocyclic group fused to a phenyl ring, in particular those wherein an aryl group is fused to a heterocyclyl or heteroaryl group. Examples of such fused ring systems are groups in which a phenyl ring is fused to a thienyl group or to a tetrahydrofuranyl group to form a benzothienyl or dihydrobenzofuranyl group. Further examples of such fused rings are groups in which a phenyl ring is fused to a dioxanyl group, a pyrrolyl group or a 2,3-dihydroinden-1-one group to form a benzodioxinyl, indolyl or a 9H-fluoren-9-one group. Most preferably, however, an aryl group, as used herein, is not fused to a monocyclic carbocyclyl, heterocyclyl or heteroaryl group or to a said fused group.

As used herein, a carbocyclyl group is a non-aromatic saturated or unsaturated monocyclic hydrocarbon ring, typically having from 3 to 6 carbon atoms. Preferably it is a saturated hydrocarbon ring (i.e. a cycloalkyl group) having from 3 to 6 carbon atoms. Examples include cyclopropyl, cyclobutyl, cyclopentyl and cyclohexyl. It is preferably cyclopropyl, cyclopentyl or cyclohexyl, most preferably cyclopropyl. A cycloalkyl group may be unsubstituted or substituted at any position. Typically, it carries 0, 1, 2 or 3 substituents.

Suitable substituents on a carbocyclyl group include halogen, C_{1-6} alkyl, C_{2-7} acyl, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, nitro, cyano, carbamoyl, mono(C_{1-6} alkyl)carbamoyl, di(C_{1-6} alkyl)carbamoyl, amino, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, oxo, $-CO_2R'$, $-CONR'R''$, $-S(O)R'$, $-S(O)_2R'$, $-S(O)NR'R''$, $-S(O)_2NR'R''$, $-NH-S(O)_2R'$ or $-NH-CO-R'$, wherein each R' and R'' is the same or different and represents hydrogen or C_{1-6} alkyl.

Preferred substituents on an carbocyclyl group include halogen, C_{1-6} alkyl, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, nitro, cyano and oxo. Particularly preferred substituents include fluorine,

chlorine, bromine, C_{1-4} alkyl, C_{1-4} alkoxy, C_{1-4} haloalkyl, nitro and oxo. Most preferably, a carbocyclyl group is unsubstituted.

As used herein, a heterocyclyl group is a non-aromatic saturated or unsaturated carbocyclic ring, typically having from 5 to 10 carbon atoms, in which one or more, for example 1, 2 or 3, of the carbon atoms is replaced by a heteroatom selected from N, O and S. Saturated heterocyclyl groups are preferred. Examples include tetrahydrofuranyl, tetrahydrothienyl, pyrrolidinyl, imidazolidinyl, pyrazolidinyl, dioxolanyl, thiazolidinyl, tetrahydropyranyl, piperidinyl, dioxanyl, piperazinyl, morpholinyl, thiomorpholinyl and thioxanyl. Further examples include dithiolanyl, oxazolidinyl, tetrahydrothiopyranyl and dithianyl. Piperazinyl, piperidinyl, thiomorpholinyl, imidazolidinyl and morpholinyl groups are preferred.

As used herein, references to a heterocyclyl group include fused ring systems in which a heterocyclyl group is fused to a phenyl group. Preferred such fused ring systems are those wherein a 5- to 6-membered heterocyclyl group is fused to a phenyl group. An example of such a fused ring system is a group wherein a 1H-imidazol-2(3*H*)-onyl group or a imidazolidin-2-onyl group is fused to a phenyl ring or a pyridine ring, to form, for example, a 1*H*-benzo[*d*]imidazol-2(3*H*)-onyl group or a 1H-imidazo[4,5-b]pyridin-2(3H)-one group. Most preferably, however, a heterocyclyl group is monocyclic.

A heterocyclic group may be unsubstituted or substituted at any position. Typically, it carries 0, 1 or 2 substituents.

Suitable substituents on a heterocyclyl group include halogen, C_{1-6} alkyl, C_{2-7} acyl, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, nitro, cyano, carbamoyl, mono(C_{1-6} alkyl)carbamoyl, di(C_{1-6} alkyl)carbomyl, amino, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, oxo, $-CO_2R'$, $-CONR'R''$, $-S(O)R'$, $-S(O)_2R'$, $-S(O)NR'R''$, $-S(O)_2NR'R''$, $-NH-S(O)_2R'$ or $-NH-CO-R'$, wherein each R' and R'' is the same or different and represents hydrogen or C_{1-6} alkyl.

Preferred substituents on a heterocyclyl group include halogen, C_{1-6} alkyl, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, nitro, cyano and oxo. Particularly preferred substituents include fluorine, chlorine, bromine, C_{1-4} alkyl, C_{1-4} alkoxy, C_{1-4} haloalkyl, nitro and oxo. Most preferably, a

heterocyclyl group is unsubstituted or substituted by one or two C_{1-2} alkyl or oxo groups. An example of a substituted heterocyclic group is S,S-dioxothiomorpholino.

As used herein, a halogen is typically chlorine, fluorine, bromine or iodine. It is preferably chlorine, fluorine or bromine. It is more preferably chlorine or fluorine.

As used herein, an alkoxy group is typically a said alkyl group attached to an oxygen atom. An alkylthio group is typically a said alkyl group attached to a thio group. A haloalkyl or haloalkoxy group is typically a said alkyl or alkoxy group substituted by one or more said halogen atoms. Typically, it is substituted by 1, 2 or 3 said halogen atoms. Preferred haloalkyl and haloalkoxy groups include perhaloalkyl and perhaloalkoxy groups such as $-CX_3$ and $-OCX_3$ wherein X is a said halogen atom, for example chlorine or fluorine. Particularly preferred haloalkyl groups are $-CF_3$ and $-CCl_3$. Particularly preferred haloalkoxy groups are $-OCF_3$ and $-OCCl_3$.

As used herein, a heteroaryl group is typically a 5- to 10-membered aromatic ring, such as a 5- or 6-membered ring, containing at least one heteroatom, for example 1, 2 or 3 heteroatoms, selected from O, S and N. Examples include pyridyl, pyrazinyl, pyrimidinyl, pyridazinyl, furanyl, thienyl, pyrazolidinyl, pyrrolyl, oxadiazolyl, isoxazolyl, thiadiazolyl, thiazolyl, imidazolyl and pyrazolyl groups. Further examples include oxazolyl and isothiazolyl. Preferred heteroaryl groups are pyridyl, thienyl, oxazolyl, isoxazolyl, thiazolyl, furanyl and pyrazolyl.

As used herein, references to a heteroaryl group include fused ring systems in which a heteroaryl group is fused to a phenyl group or to a monocyclic heterocyclyl group. Preferred such fused ring systems are those wherein a 5- to 6-membered heteroaryl group is fused to a phenyl group or to a 5- to 6- membered heterocyclyl group. Examples of such fused ring systems are benzofuranyl, benzothiophenyl, indolyl, benzimidazolyl, benzoxazolyl, quinolinyl, quinazolinyl, isoquinolinyl and 1H-imidazo[4,5-b]pyridin-2(3H)-one moieties. Most preferably, said fused ring system is a 1H-imidazo[4,5-b]pyridin-2(3H)-one moiety.

A heteroaryl group may be unsubstituted or substituted at any position. Typically, it carries 0, 1, 2 or 3 substituents.

Suitable substituents on a heteroaryl group include halogen, C_{1-6} alkyl, C_{2-7} acyl, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, nitro, cyano, carbamoyl, mono(C_{1-6} alkyl)carbamoyl, di(C_{1-6} alkyl)carbamoyl, amino, mono(C_{1-6} alkyl)amino, di(C_{1-6}

alkyl)amino, $-CO_2R'$, $-CONR'R''$, $-S(O)R'$, $-S(O)_2R'$, $-S(O)NR'R''$,$-S(O)_2NR'R''$, $-NH-S(O)_2R'$ or $-NH-CO-R'$, wherein each R' and R'' is the same or different and represents hydrogen or C_{1-6} alkyl.

Preferred substituents on a heteroaryl group include halogen, C_{1-6} alkyl, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl, C_{1-6} haloalkoxy, mono(C_{1-6} alkyl)amino, di(C_{1-6} alkyl)amino, nitro and cyano. Particularly preferred substituents include fluorine, chlorine, bromine, C_{1-4} alkyl, C_{1-4} alkoxy, C_{1-4} haloalkyl and nitro. Most preferred substituents include fluorine, chlorine, bromine, C_{1-2} alkyl and C_{1-2} haloalkyl substituents.

When R^1 in the formula (V) is an aryl or heteroaryl group it is typically unsubstituted or substituted by one, two or three substituents selected from halogen, C_{1-6} alkyl, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} haloalkyl or C_{1-6} haloalkoxy. Preferably, it is unsubstituted or substituted by one or two substituents selected from fluorine, chlorine, bromine, C_{1-4} alkyl, C_{1-4} alkoxy, C_{1-4} alkylthio, C_{1-4} haloalkyl or C_{1-4} haloalkoxy. More preferably, it is unsubstituted or substituted by a single fluorine, chlorine, C_{1-2} alkyl, C_{1-2} alkoxy, C_{1-2} alkylthio, C_{1-2} haloalkyl or C_{1-2} haloalkoxy substituent.

Typically, R^1 in formula (V) is C_{1-6} alkyl or aryl. Preferably, R^1 is C_{1-2} alkyl or aryl. More preferably, R^1 is C_{1-2} alkyl or phenyl. More preferably, R^1 is an unsubstituted phenyl group.

Typically, R^2 in formula (V) is hydrogen or C_{1-4} alkyl. Preferably, R^2 is hydrogen.

Typically, R^3 in formula (V) is halogen, hydroxy, C_{1-4} alkyl, C_{1-4} alkoxy, C_{1-4} alkylthio, C_{1-4} haloalkyl, C_{1-4} haloalkoxy, amino, mono(C_{1-4} alkyl)amino or di(C_{1-4} alkyl)amino. Preferably, R^3 is fluorine, chlorine, bromine, C_{1-2} alkyl, C_{1-2} alkoxy, C_{1-2} alkylthio, C_{1-2} haloalkyl, C_{1-2} haloalkoxy, amino, mono(C_{1-2} alkyl)amino or di(C_{1-2} alkyl) amino. More preferably, R^3 is methyl, trifluoromethyl, fluorine, chlorine or bromine. Most preferably, R^3 is methyl or chlorine.

Typically, n in formula (V) is 0, 1 or 2. Preferably, n is 0 or 1. Most preferably, n is 0.

Typically, R^4 in formula (V) is hydrogen or C_{1-4} alkyl. Preferably, R^4 is hydrogen or C_{1-2} alkyl. More preferably, R^4 is hydrogen or methyl. Most preferably, R^4 is hydrogen

Typically, X in formula (V) is -CO-, $-S(O)_2-$ or $-CO-NR'-$, wherein R' represents hydrogen or a C_1-C_2 alkyl group. Preferably, X is -CO- or $-CO-NR'-$.

When R^5 in formula (V) is a heterocyclyl or heterocyclyl group which is substituted by a C_1-C_6 hydroxyalkyl group or a -(C_1-C_4 alkyl)-X_1-(C_1-C_4 alkyl)-X_2-(C_1-C_4 alkyl) group, the heterocyclyl or heteroaryl group is typically a 5- or 6- membered ring. Preferably, it is a 5- or 6- membered heteroaryl group, for example a furanyl group.

Typically, the C_1-C_6 hydroxyalkyl group in formula (V) is a -CH_2-OH group. Typically, X_1 in the formula (V) is -NR'-, wherein R' is hydrogen or C_1-C_2 alkyl. Typically, X_2 in formula (V) is -$S(O)_2$-.

Typically, A_1 in formula (V) is an aryl or heteroaryl group. Preferably, A_1 is a monocyclic aryl or heteroaryl group, a naphthyl group or a heteroaryl group fused to a monocyclic oxo substituted heterocyclyl group. More preferably, A_1 is a phenyl group, a monocyclic 5- or 6- membered heteroaryl group or a 5- to 6- membered heteroaryl group fused to a monocyclic oxo substituted 5- to 6- membered heterocyclyl group (for example an oxo substituted imidazolidine group). Most preferably, A_1 is a phenyl, pyridyl, furanyl, thiazolyl, oxazolyl, isoxazolyl, thienyl or 1H-imidazo[4,5-b]pyridin-2-(3H)-one moiety.

Typically, the moiety A_1 in formula (V) is unsubstituted or substituted by 1 or 2 substituents selected from halogen, cyano, nitro, C_1-C_4 alkyl, C_1-C_4 haloalkyl and C_1-C_4 alkoxy substituents. Preferably, the substituents are selected from halogen, cyano, C_1-C_2 alkyl, C_1-C_2 haloalkyl and C_1-C_2 alkoxy substituents.

Typically, Y in formula (V) represents a direct bond, a C_1-C_2 alkylene group, -SO_2- or -O-.

Typically, A_2 in formula (V) is a phenyl, 5- to 6- membered heteroaryl, 5- to 6- membered heterocyclyl or C_3-C_6 cycloalkyl group. Preferably, A_2 is a piperazinyl, pyridyl, morpholinyl, thiomorpholinyl, pyrrolidinyl, piperidinyl, pyrazinyl, cyclopropyl or phenyl group.

Typically, when A_2 in formula (V) is a heterocyclyl group it is attached to the moiety Y via a N atom.

Typically, the moiety A_2 in formula (V) is unsubstituted or substituted by one or two substituents which are selected from C_1-C_4 alkyl and halogen substituents when A_2 is a heteroaryl or aryl group and which are selected from C_1-C_4 alkyl, halogen and oxo substituents when A_2 is a carbocyclic or heterocyclyl group.

Most preferably, A_2 in formula (V) is a piperazinyl, pyridyl, morpholinyl, pyrrolidinyl, piperidinyl, pyrazinyl, cyclopropyl, phenyl or S,S-dioxo-thiomorpholino group, which group is unsubstituted or is substituted by a C_1-C_2 alkyl group.

Preferred compounds of formula (V) are those in which:

- R^1 is C_{1-6} alkyl or aryl;
- R^2 is hydrogen or C_{1-4} alkyl;
- R^3 is halogen, hydroxy, C_{1-4} alkyl, C_{1-4} alkoxy, C_{1-4} alkylthio, C_{1-4} haloalkyl, C_{1-4} haloalkoxy, amino, mono(C_{1-4} alkyl)amino or di(C_{1-4} alkyl)amino or, preferably, R^3 is fluorine, chlorine, bromine, C_{1-2} alkyl, C_{1-2} alkoxy, C_{1-2} alkylthio, C_{1-2} haloalkyl, C_{1-2} haloalkoxy, amino, mono(C_{1-2} alkyl)amino or di (C_{1-2} alkyl)amino;
- n is 0, 1 or 2;
- R^4 is hydrogen or C_{1-4} alkyl;
- X is -CO-, -CO-NR′ or -$S(O)_2$-, wherein R′ is hydrogen or a C_1-C_2 alkyl group; and
- R^5 is a 5- or 6- membered heterocyclyl or heteroaryl ring which is substituted by a C_1-C_6 hydroxyalkyl group or a -(C_1-C_4 alkyl)-X_1-(C_1-C_4 alkyl)-X_2-(C_1-C_4 alkyl) group, wherein X_1 and X_2 are as defined above, or R^5 represents -A_1-Y-A_2, wherein:
  - A_1 is an aryl or heteroaryl group;
  - Y is a direct bond, a C_1-C_2 alkylene group, -SO_2- or -O-; and
  - A_2 is an aryl, heteroaryl, heterocyclyl or carbocyclyl group,

the aryl moiety in the R^1 group being unsubstituted or substituted by 1, 2 or 3 substituents selected from halogen, C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 alkylthio, C_1-C_6 haloalkyl and C_1-C_6 haloalkoxy groups,

the A_1 moiety being unsubstituted or substituted by 1 or 2 substituents selected from halogen, cyano, nitro, C_1-C_4 alkyl, C_1-C_4 haloalkyl and C_1-C_4 alkoxy substituents; and

the A_2 moiety being unsubstituted or substituted by one or two substituents which are selected from C_1-C_4 alkyl and halogen substituents when A_2 is a heteroaryl or aryl group and which are selected from C_1-C_4 alkyl, halogen and oxo substituents when A_2 is a carbocyclic or heterocyclyl group.

Further preferred compounds of formula (V) are those wherein:

- R^1 is C_{1-2} alkyl or phenyl;

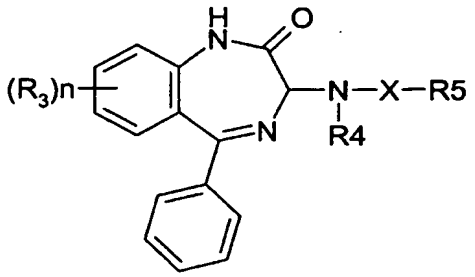
- R^2 is hydrogen or C_{1-4} alkyl;
- R^3 is methyl, trifluoromethyl, fluorine, chlorine or bromine;
- n is 0 or 1;
- R^4 is hydrogen or C_{1-2} alkyl;
- X is -CO-, -CO-NR′- or -S(O)$_2$, wherein R′ is hydrogen or a C_1-C_2 alkyl group; and
- R^5 is a 5- or 6- membered heterocyclyl or heteroaryl group which is substituted by a C_1-C_6 hydroxyalkyl group or a -(C_1-C_4 alkyl)-NR′-(C_1-C_4 alkyl)-SO_2-(C_1-C_4 alkyl) group, wherein R′ is hydrogen or C_1-C_2 alkyl, or R^5 represents -A_1-Y-A_2, wherein:
- A_1 is a phenyl group, a monocyclic 5- or 6- membered heteroaryl group or a 5- or 6- membered heteroaryl group fused to a monocyclic oxo-substituted 5- to 6- membered heterocyclyl group;
- Y represents a direct bond, a C_1-C_2 alkylene moiety, -SO_2- or -O-; and
- A_2 is a phenyl, 5- to 6- membered heteroaryl, 5- to 6- membered heterocyclyl or C_3-C_6 cycloalkyl group,

the phenyl moiety in the R^1 group being unsubstituted or substituted by one or two substituents selected from fluorine, chlorine, bromine, C_{1-4} alkyl, C_{1-4} alkoxy, C_{1-4} alkylthio, C_{1-4} haloalkyl or C_{1-4} haloalkoxy;

the A_1 moiety being unsubstituted or substituted by 1 or 2 substituents selected from halogen, cyano, nitro, C_1-C_4 alkyl, C_1-C_4 haloalkyl and C_1-C_4 alkoxy substituents; and

the A_2 moiety being unsubstituted or substituted by 1 or 2 substituents which are selected from C_1-C_4 alkyl, halogen and oxo substituents when A_2 is a heterocyclyl or cycloalkyl group and which are selected from C_1-C_4 alkyl and halogen substituents when A_2 is a phenyl or heteroaryl group.

Particularly preferred compounds of the invention are compounds of formula (Va) and pharmaceutically acceptable salts thereof



(Va)

wherein:

- X is -CO- or -CO-NH-; and
- R^5 is a 5- to 6- membered heteroaryl group, for example a furanyl group, which is substituted by $-CH_2-OH$ or $-(C_1-C_4$ alkyl$)-N(CH_3)-(C_1-C_4$ alkyl$)-SO_2-(C_1-C_4$ alkyl) or R_5 represents $-A_1-Y-A_2$, wherein:
- A_1 is a phenyl, pyridyl, furanyl, thiazolyl, oxazolyl, isoxazolyl, thienyl or 1H-imidazo[4,5-b]pyridin-2-(3H)-one moiety, which is unsubstituted or substituted by 1 or 2 substituents selected from halogen, cyano, C_1-C_2 alkyl, C_1-C_2 haloalkyl and C_1-C_2 alkoxy substituents;
- Y is a direct bond, a C_1-C_2 alkylene group, $-SO_2-$ or -O-; and
- A_2 is a piperazinyl, pyridyl, morpholinyl, pyrrolidinyl, piperidinyl, pyrazinyl, cyclopropyl, phenyl or S,S-dioxo-thiomorpholino group, which is unsubstituted or substituted by a C_1-C_2 alkyl group.

In the compounds of formula (Va), typically n is 0 and R_4 is hydrogen. Preferably, in the compounds of formula (Va), A_1 is a phenyl or furanyl group which is unsubstituted or substituted by a chlorine atom. Preferably, Y is a direct bond or a methylene group. Preferably, A_2 is a morpholino or S,S-dioxo-thiomorpholino group.

Compounds of the formula (V) containing one or more chiral centre may be used in enantiomerically or diasteroisomerically pure form, or in the form of a mixture of isomers. For the avoidance of doubt, the chemical structures depicted herein are intended to embrace all stereoisomers of the compounds shown, including racemic and non-racemic mixtures and pure enantiomers and/or diastereoisomers.

Preferred compounds of formula (V) are optically active isomers. Thus, for example, preferred compounds of formula (V) containing only one chiral centre include an R enantiomer in substantially pure form, an S enantiomer in substantially pure form and

enantiomeric mixtures which contain an excess of the R enantiomer or an excess of the S enantiomer. For the avoidance of doubt, the compounds of the formula (V) can, if desired, be used in the form of solvates.

As used herein, a pharmaceutically acceptable salt is a salt with a pharmaceutically acceptable acid or base. Pharmaceutically acceptable acids include both inorganic acids such as hydrochloric, sulphuric, phosphoric, diphosphoric, hydrobromic or nitric acid and organic acids such as citric, fumaric, maleic, malic, ascorbic, succinic, tartaric, benzoic, acetic, methanesulphonic, ethanesulphonic, benzenesulphonic or p-toluenesulphonic acid. Pharmaceutical acceptable bases include alkali metal (e.g. sodium or potassium) and alkaline earth metal (e.g. calcium or magnesium) hydroxides and organic bases such as alkyl amines, aralkyl amines or heterocyclic amines.

Particularly preferred compounds of formula (V) include:

6-(4-Methyl-piperazin-1-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4] diazepin-3-yl)-nicotinamide;

3,4,5,6-Tetrahydro-2H-[1,2']bipyridinyl-5'-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl-benzamide;

(S)-2-Chloro-4-morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4] diazepin-3-yl)-benzamide;

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-4-fluoro-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl-benzamide;

(S)-5-Chloro-2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-5-fluoro-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-5-(4-Methyl-piperazin-1-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-5-Pyrrolidin-1-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-5-Piperidin-1-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-5-Dimethylaminomethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-4-Fluoro-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-piperidin-1-yl-benzamide;

(S)-4-Fluoro-2-morpholino-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4] diazepin-3-yl)-benzamide;

(S)-4-Cyano-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-pyrrolidin-1-yl-benzamide;

(S)-4-Cyano-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-piperidine-1-yl-benzamide;

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-pyrrolidin-1-yl-4-trifluoromethyl-benzamide;

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-piperidin-1-yl-4-trifluoromethyl-benzamide;

(S)-2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-4-trifluoromethyl-benzamide;

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-pyrrolidin-1-yl-5-trifluoromethyl-benzamide;

(S)-2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-5-trifluoromethyl-benzamide;

(S)-2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide;

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide;

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-2-methyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-4-methyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-6-methyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-2-Chloro-6-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-3-Cyclopropyl-2-oxo-2,3-dihydro-imidazo[4,5-b]pyridine-1-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-3-(4-Methyl-piperazine-1-sulfonyl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-4-(4-Methyl-piperazin-1-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4] diazepin-3-yl)-benzamide;

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-3-(piperidine-1-sulfonyl)-benzamide;

(S)-3-(Morpholine-4-sulfonyl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4] diazepin-3-yl)-benzamide;

(S)-5-Morpholin-4-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-5-Hydroxymethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-5-(1,1-Dioxo-1λ6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-2-Chloro-4-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-2-Chloro-5-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-5-{[(2-Methanesulfonyl-ethyl)-methyl-amino]-methyl}-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl-amide;

(S)-2-Pyridin-3-yl-thiazole-4-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-2-Pyridin-4-yl-thiazole-4-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-4-Methyl-2-pyrazin-2-yl-thiazole-5-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-2-Morpholin-4-ylmethyl-furan-3-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-3-Morpholin-4-ylmethyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4] diazepin-3-yl)-benzamide;

(S)-5-Morpholin-4-ylmethyl-isoxazole-3-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-3-Morpholin-4-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-5-Pyridin-2-yl-thiophene-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-2-Methyl-4-(morpholin-4-sulfonyl)-furan-3-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-6-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide;

(S)-3-Morpholin-4-ylmethyl-thiophene-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

(S)-5-Morpholin-4-ylmethyl-thiophene-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide;

(S)-5-Phenyl-oxazole-4 carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide;

1-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-3-(4-phenoxy-phenyl)-urea;

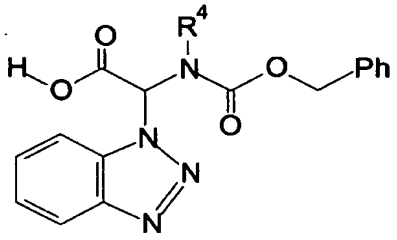
an N-oxide of any of the above compounds;

and pharmaceutically acceptable salts thereof.

The compounds of formulae (I), (II), (III) and (IV) are known compounds. They are disclosed, for example, in WO 00/195910, WO 00/004900, WO 03/053344, US-A-4324794 and WO 01/00612, and can be prepared by the processes set out in those documents.

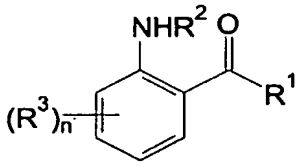
WO 00/195910, WO 00/004900, WO 03/053344, US-A-4324794 and WO 01/00612 are incorporated herein by reference. Any of the compounds disclosed as fusion inhibitors in those documents can be used in the present invention.

Compounds of formula (V) may be prepared by reacting glyoxylic acid (HCO-CO_2H), benzotriazole and an appropriate benzyl carbamate at reflux in toluene, under Dean-Stark conditions giving the key protected amino acid of formula (II′)



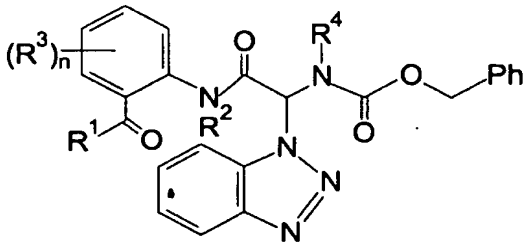
(II′)

The thus obtained amino acid of formula (II′) can then be reacted with a suitable chlorinating agent, such as oxalyl chloride, followed by reaction with a 2-aminobenzophenone of formula (III′)



(III′)

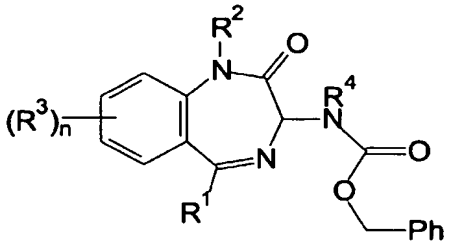
to give the intermediate amide of formula (IV′)



(IV′)

which need not be characterized.

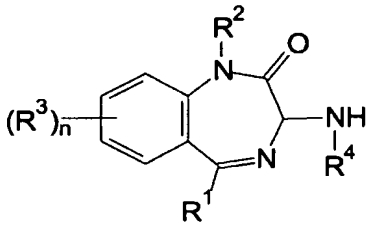
The compound of formula (IV′) can then be subjected to ammonolysis followed by



(V′)

ring closure in acetic acid containing ammonium acetate to obtain the protected benzodiazepine of formula (V′)

The compound of formula (V′) can then be deprotected using hydrogen bromide in acetic acid to yield the deprotected amine of formula (VI′).



(VI′)

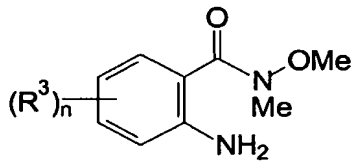
Compounds of formula (V), in which X is -CO- or -CO-NR′ can be prepared by reacting a compound of formula (VI′), as defined above, with an acid anhydride in a suitable solvent, preferably pyridine at ambient temperature, or with an acid chloride in a suitable solvent in the presence of a base, preferably in THF at ambient temperature with triethylamine present. Alternatively, the compounds can be produced by reaction of a compound of formula (VI′) with an acid in a suitable solvent in the presence of a base and a coupling agent, preferably in THF at ambient temperature with triethylamine and *O*-benzotriazol-1-yl-N, N, N', N'-tetramethyluronium hexafluorophosphate (HBTU) present.

If the acid chloride used is an amino carbonyl chloride, the compound of formula (V) is a urea. In the case where R′ in the X moiety is hydrogen, such compounds may also be prepared by the reaction of a compound of formula (VI′) with an isocyanate. This reaction is preferably carried out in THF at ambient temperature. Alternatively, the isocyanate may be

prepared *in situ* from the relevant amine and phosgene, in the presence of a base, usually triethylamine, again in THF. Compounds in which R′ is other than hydrogen can, of course, be prepared by reacting a corresponding compound in which R′ is hydrogen with an appropriate alkylating agent, for example L-(C_1-C_6 alkyl) wherein L is a leaving group, for example chlorine.

Compounds of formula (V), in which X is -$S(O)_2$- may be prepared by the reaction of a compound of formula (VI′) with a suitable sulfonyl chloride. Similarly, compounds of formula (V), in which X is -S(O)- may be prepared by the reaction of a compound of formula (VI') with a suitable sulfinyl chloride

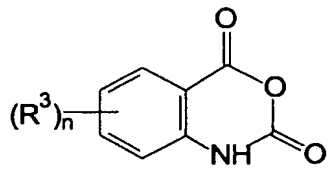
In the preparation of the benzodiazepine skeleton, commercially available aminobenzophenone compounds of formula (III′) can be used where possible. Compounds of formula (III′) which are not commercially available can be prepared by known methods, for example by reaction of a Weinreb type amide of formula (VII′)



(VII′)

with a group R^1-Li or a Grignard reagent such as R^1-MgBr. Preferably this reaction is carried out in THF at –100°C.

Compounds of formula (VII′) are known compounds or can be prepared by analogy with known methods. For example, they can be prepared from the reaction of isatoic anhydrides of formula (VIII′)



(VIII′)

with N,O-dimethyl hydroxylamine under standard reaction conditions.

The starting materials of formula (II′), (III′), (VII′), and (VIII′) are known compounds, or may be prepared by analogy with known methods.

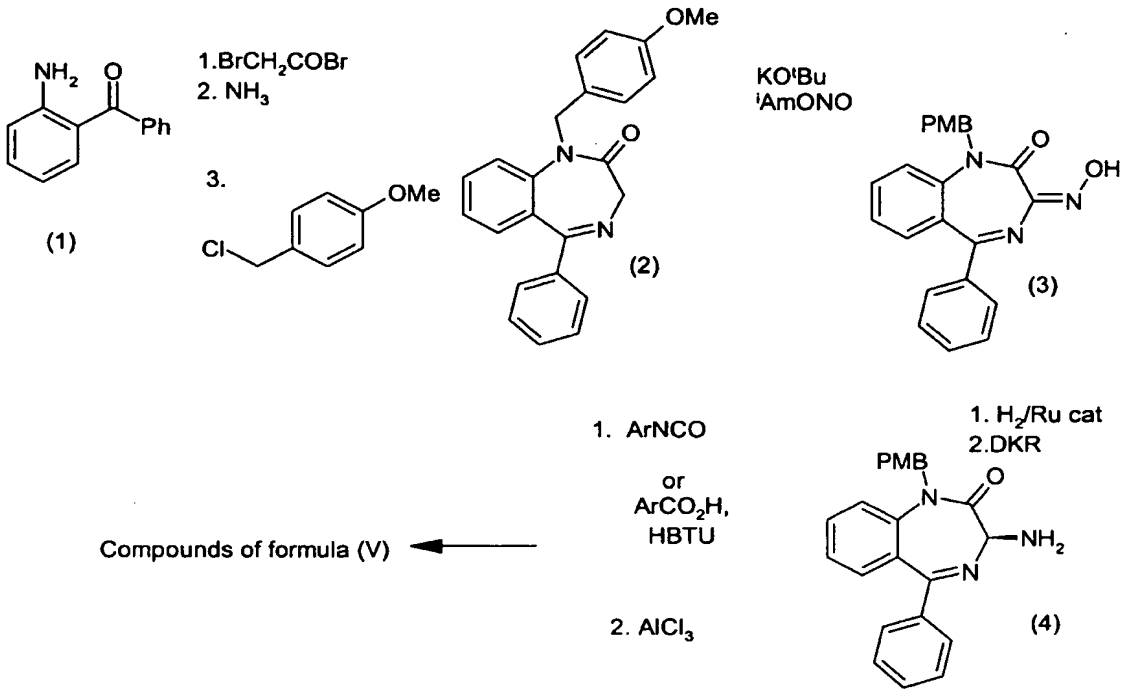
Further synthetic manipulation of the thus obtained compounds of formula (V) may be carried out by conventional methods to achieve further compounds of formula (V). The benzodiazepines of formula (V) can be salified by treatment with an appropriate acid or base.

Although the described route to the claimed compounds of formula (V) provides an adequate synthesis for laboratory scale preparations, an alternative route was sought which has potential as a manufacturing route. The same starting material (2-amino-benzophenone) (1) is used in both, however in the alternative route, the benzodiazepine ring system is formed by reaction initially with bromoacetyl bromide (or an equivalent reagent) followed by ring closure with ammonia. These reactions are carried out in a suitable solvent, such as dichloromethane, and at a suitable temperature which may range from -20 to 150°C. In order to protect the NH functionality, at this stage the unsubstituted benzodiazepine is reacted with a base, and an alkylating agent. For instance sodium hydride in DMF followed by addition of 4-methoxy-benzyl chloride gives rise to the intermediate (2) shown below. Further reaction of this material with a base (e.g. potassium tert-butoxide) in a suitable solvent (e.g. THF or DMF) followed by quenching with isoamyl nitrite (or an alternative similar reagent) furnishes the oxime intermediate (3) which may be converted into the racemic primary amine by methods which include the use of hydrogen and a suitable catalyst. This amine then undergoes a Dynamic Kinetic Resolution (DKR) procedure by which the racemic amine in the presence of a suitable optically active acid, and a suitable aldehyde gives rise to precipitation of the salt of the desired (S)-amine (4) in good yield and exceptionally high enantiomeric excess. A suitable acid for this conversion can be e.g. Camphorsulfonic acid, Boc-phenyl alanine or the like, and a suitable aldehyde may be a benzaldehyde such as 3,5-dichloro salicylaldehyde.

The optically amine thus formed may then be transformed into a desired derivative, such as an amide or urea. The amide formations may be carried out using a suitable carboxylic acid and a coupling reagent, or a carbonyl chloride or other suitable reagent, and the ureas prepared using either a suitable isocyanate, or alternatively reaction with phosgene followed by a suitable amine.

These derivatives thus formed may then have the protecting group removed. This may be carried out in the presence of a Lewis Acid, such as aluminium chloride, boron trifluoride, titanium tetrachloride, or the like. These reactions are carried out in a suitable

inert solvent, such as dichloromethane. Reaction temperatures may range from -20 to 150°C, but are typically carried out at room temperature or below.



In a particularly preferred embodiment of the invention, component (a) is 1-cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one, 2-[2-(1,2-dihydro-benzotriazol-1-ylmethyl)-benzoimidazol-1-yl]]ethyl}-diethyl-amine, {2-[2-(3-iodo-2,3-dihydro-indazol-1-ylmethyl)-benzimidazol-1-yl]-ethyl}-dimethyl-amine or a pharmaceutically acceptable salt thereof and component (b) is (S)-2-Chloro-4-morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide or 5-(1,1-dioxo-1λ6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)amide or a pharmaceutically acceptable salt thereof.

The present invention also provides a pharmaceutical composition according to the invention, for use in the treatment of the human or animal body. Also provided is the use of (a) a said RSV fusion protein inhibitor and (b) a said benzodiazepine derivative, in the manufacture of a medicament for use in treating or preventing an RSV infection.

The present invention also provides a method of treating or preventing an RSV infection in a patient, which method comprises the administration to said patient of (a) a said RSV fusion protein inhibitor and (b) a said benzodiazepine derivative.

Typically, the amount of component (a) in the composition of the invention is from 0.025 wt% to 10 wt%, preferably from 0.25 wt% to 5 wt%, more preferably from 1 wt% to 3.5 wt%, for example about 2.5 wt%, based on the total weight of the composition.

Typically, the amount of component (b) in the composition of the invention is from 0.025 wt% to 10 wt%, preferably from 0.25 wt% to 5 wt%, more preferably from 1 wt% to 3.5 wt%, for example about 2.5 wt%, based on the total weight of the composition.

Typically, the total amount of components (a) and (b) in the composition of the invention is from 0.05 to 20 wt%, preferably from 0.5 to 10 wt%, more preferably from 2 to 7 wt%, for example about 5 wt%, based on the total weight of the composition.

RSV is prevalent among children younger than two years of age, adults suffering from asthma, chronic obstructive pulmonary disorder (COPD) or immunodeficiency and the elderly. It is a particularly serious risk amongst children who suffer from chronic lung disease. Accordingly, the said composition or medicament is typically for use in treating a patient who is a child under two years of age, patients with asthma, COPD or immunodeficiency the elderly or persons in long term care facilities. Typically, said child suffers from chronic lung disease.

Further, anti-RSV prophylaxis is recommended for infants born at 32 weeks of gestation or earlier, until they reach 6 months of age, the elderly, persons with immunedeficiency and those in long term care facilities. Accordingly, the said composition or medicament is typically for use in preventing RSV infection in an infant less than 6 years of age, who was born after 32 weeks of gestation or less, the elderly, persons with immunosufficiency and those in long term care facilities.

As described above, RSV strains upon exposure to fusion inhibitors known in the art rapidly develop resistance. In order to minimize the risk of development of resistance to

fusion inhibitors it is desirable to combine them with another inhibitor of RSV replication with a different mode of action. To our knowledge, the benzodiazepine derivatives disclosed above are the first class of compounds with a novel mode of action. Accordingly, the compositions of the invention are characterized by a very low resistance profile, which makes them particularly suitable for therapeutic and prophylactic applications.

The present invention also covers situations where components (a) and (b) are administered separately. Thus, for example, component (a) can be administered up to 24 hours before component (b). Alternatively, component (b) can be administered up to 24 hours before component (a). More usually, when components (a) and (b) are administered separately, they are administered within 12 hours, preferably within 6 hours, of each other.

The present invention therefore also provides a product comprising (a) a said RSV fusion protein inhibitor and (b) a said benzodiazepine derivative for separate, simultaneous or sequential use in the treatment of the human or animal body. Typically, said product is for separate, simultaneous or sequential use in treating or preventing an RSV infection.

Also provided is the use of a said RSV fusion protein inhibitor in the manufacture of a medicament for use in treating or preventing an RSV infection by co-administration with a said benzodiazepine derivative. The present invention also provides the use of a said benzodiazepine derivative in the manufacture of a medicament for use in treating or preventing an RSV infection, by co-administration with a said RSV fusion protein inhibitor.

When components (a) and (b) are administered separately, they are typically formulated as described above. The amount of active ingredient in each separate formulation will, of course, correspond to the amount of component (a) or (b) given above for the combined formulation. Thus, when components (a) and (b) are administered separately, a first formulation is typically provided which contains from 0.025 wt% to 10 wt%, preferably from 0.25 wt% to 5 wt%, more preferably from 1 wt% to 3.5 wt%, for example about 2.5 wt%, of a said RSV fusion protein inhibitor, based on the total weight of the formulation. Similarly, a second formulation is typically provided which contains from 0.025 wt% to 10 wt%, preferably from 0.25 wt% to 5 wt%, more preferably from 1 wt% to 3.5 wt%, for example around 2.5 wt%, of a said benzodiazepine derivative, based on the total weight of the formulation. The two formulations can be administered separately in any order.

Preferably, the compositions and medicaments of the invention have an activity greater than the combined individual activities of compounds (a) and (b). Thus, components (a) and (b) typically interact synergistically. Preferably, therefore, in the formulations and the medicaments of the invention, component (a) and component (b) are each present in an amount producing a synergistic therapeutic effect in treating or preventing an RSV infection.

The anti-RSV compositions of the invention may be administered in a variety of dosage forms. Thus, they can be administered orally, for example as tablets, troches, lozenges, aqueous or oily suspensions, dispersible powders or granules. The compounds of the invention may also be administered parenterally, whether subcutaneously, intravenously, intramuscularly, intrasternally, transdermally or by infusion techniques. The compounds may also be administered as suppositories.

In a preferred embodiment, administration is by intravenous, intranasal or intrabronchial means. In particular, formulations for treating or preventing RSV can advantageously be administered intranasally. The present invention therefore also provides an inhaler or nebuliser containing a medicament which comprises (i) a composition of the invention comprising component (a) and component (b), as defined above, and (ii) a pharmaceutically acceptable carrier or diluent.

The anti-RSV compositions of the invention are typically formulated for administration with a pharmaceutically acceptable carrier or diluent. For example, solid oral forms may contain, together with the active compound(s), diluents, e.g. lactose, dextrose, saccharose, cellulose, corn starch or potato starch; lubricants, e.g. silica, talc, stearic acid, magnesium or calcium stearate, and/or polyethylene glycols; binding agents; e.g. starches, arabic gums, gelatin, methylcellulose, carboxymethylcellulose or polyvinyl pyrrolidone; disaggregating agents, e.g. starch, alginic acid, alginates or sodium starch glycolate; effervescing mixtures; dyestuffs; sweeteners; wetting agents, such as lecithin, polysorbates, laurylsulphates; and, in general, non toxic and pharmacologically inactive substances used in pharmaceutical formulations. Such pharmaceutical preparations may be manufactured in known manner, for example, by means of mixing, granulating, tableting, sugar coating, or film coating processes.

Liquid dispersions for oral administration may be syrups, emulsions and suspensions. The syrups may contain as carriers, for example, saccharose or saccharose with glycerine and/or mannitol and/or sorbitol.

Suspensions and emulsions may contain as carrier, for example a natural gum, agar, sodium alginate, pectin, methylcellulose, carboxymethylcellulose, or polyvinyl alcohol. The suspension or solutions for intramuscular injections may contain, together with the active compound, a pharmaceutically acceptable carrier, e.g. sterile water, olive oil, ethyl oleate, glycols, e.g. propylene glycol, and if desired, a suitable amount of lidocaine hydrochloride.

Solutions for injection or infusion may contain as carrier, for example, sterile water or preferably they may be in the form of sterile, aqueous, isotonic saline solutions.

Preferably, the anti-RSV compositions of the invention are solubilised in a carrier containing (a) a pharmaceutically acceptable oil selected from esterification or polyether products of glycerides with vegetable oil fatty acids of chain length C_8-C_{10} and (b) a pharmaceutically acceptable surfactant selected from oleate and laurate esters of a polyalcohol copolymerized with ethylene oxide. Particularly preferred carriers contain Labrafil as the oil and Tween 20 or Tween 80 as the surfactant.

The anti-RSV compositions of the invention may also be suspended in PEG 400 for oral administration.

A therapeutically effective amount of an anti-RSV composition of the invention is administered to a patient. A typical dose is from about 0.001 to 50 mg, typically 0.5 to 30 mg, preferably 1 to 20 mg active ingredient per kg of body weight, according to the activity of the specific composition, the age, weight and conditions of the subject to be treated, the type and severity of the disease and the frequency and route of administration. Preferably, daily dosage levels are from 5 mg to 2 g active ingredient.

The following Examples illustrate the invention. They do not however, limit the invention in any way. In this regard, it is important to understand that the particular assays used in the Examples section are designed only to provide an indication of antiviral activity. There are many assays available to determine the activity of given compounds against RSV, and a negative result in any one particular assay is therefore not determinative.

EXAMPLES

Intermediate 1

2-Chloro-4-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid

A mixture of 4-amino-2-chlorobenzoic acid (172mg) and ethenesulfonyl-ethene (0.15ml) in water (3ml) containing sodium carbonate (212mg) was heated to 100C for 18h. The mixture was allowed to cool and was acidified with 2N HCl. The off-white precipitate was collected and dried (263mg)

LC/MS RT= 4.09mins, ES- 288,290

Intermediate 2

2-Chloro-5-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid

A mixture of 5-amino-2-chlorobenzoic acid (172mg) and ethenesulfonyl-ethene (0.15ml) in water (3ml) was heated to 100C for 18h. The mixture was allowed to cool and was extracted with dichloromethane. The dried extracts were evaporated giving a pale brown solid (265mg)

LC/MS RT= 4.13mins, ES- 288,290

Intermediate 3

2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-nicotinic acid

This material was prepared as described for Intermediate 1 except that 2-amino-nicotinic acid (138mg) was used. The title compound was isolated as an off-white solid (93mg)

Intermediate 4

2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-3-methyl-benzoic acid

This material was prepared as described for Intermediate 2 except that 2-amino-3-methyl-benzoic acid (302mg) was used. The title compound was isolated as a pale brown solid (486mg)

Intermediate 5

2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-4-methyl-benzoic acid

This material was prepared as described for Intermediate 2 except that 2-amino-4-methyl-benzoic acid (302mg) was used. The title compound was isolated as a brown solid (430mg)

Intermediate 6

2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-6-methyl-benzoic acid

This material was prepared as described for Intermediate 2 except that 2-amino-6-methyl-benzoic acid (302mg) was used. The title compound was isolated as a brown solid (490mg)

Intermediate 7

3-(4-Methyl-piperazine-1-sulfonyl)-benzoic acid

A solution of 3-chlorosulfonyl-benzoic acid (89mg) 4-dimethylamino-pyridine (catalytic amount) and N-methylpiperazine (0.045ml) in dichloromethane (10ml) was heated to reflux for 2h. The solvent was then evaporated and the crude material used without purification or characterisation in the next synthetic step.

Intermediate 8

3-Piperidine-1-sulfonyl-benzoic acid

This material was prepared as described for Intermediate 7 except that piperidine was used as the nucleophile. As for Intermediate 7 the material was used crude.

Intermediate 9

3-(Morpholine-4-sulfonyl)-benzoic acid

This material was prepared as described for Intermediate 7 except that morpholine was used as the nucleophile. As for Intermediate 7 the material was used crude.

Intermediate 10

2-Chloro-6-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid

This material was prepared as described for Intermediate 2 except that 2-amino-6-chloro-benzoic acid (343mg) was used. The title compound was isolated as a buff solid (405mg)

Intermediate 11

5-Chloro-2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid

This material was prepared as described for Intermediate 2 except that 2-amino-5-chloro-benzoic acid (200mg) was used. The title compound was isolated as a white solid (233mg)
^{1}H NMR (DMSO, δ) 3.25 (brs, 4H) 3.47 (brs, 4H) 7.31 (d, 1H) 7.54 (dd, 1H) 7.71 (d, 1H)
LC/MS RT = 4.66 min Found ES^+ = 290,292

Intermediate 12

2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-5-fluoro-benzoic acid

This material was prepared as described for Intermediate 2 except that 2-amino-5-fluoro-benzoic acid (200mg) was used. The title compound was isolated as a white solid (310mg)
^{1}H NMR (DMSO, δ) 3.28 (m, 4H) 3.42 (m, 4H) 7.33-7.56 (m, 3H)
LC/MS RT = 4.28 min Found ES$^-$ = 272

Intermediate 13

4-Fluoro-2-thiomorpholin-4-yl-benzoic acid

A mixture of 2,4-difluoro-benzoic acid (0.5g), thiomorpholine (0.33ml) and triethylamine (0.88ml) in acetonitrile (2ml) was heated to 200C in a microwave reactor for 20mins. The residue was partitioned between water and dichloromethane. The dried organic layer was evaporated and then purified on a silica gel SPE cartridge. Elution with dichloromethane followed by a gradient of dichloromethane:ethanol:0.880 ammonia; 800:8:1 to 200:8:1 gave the title material as a white solid (292mg)

^{1}H NMR (DMSO, δ) 2.81 (m, 4H) 3.27 (m, 4H) 7.11 (m, 1H) 7.40 (dd, 1H) 7.95 (m, 1H)

Intermediate 14

2-(1,1-Dioxo-4-oxy-1λ6-thiomorpholin-4-yl)-4-fluoro-benzoic acid

Intermediate 11 (262mg) and potassium peroxymonosulfate (1.34g) in methanol (5ml) and water (2.5ml) was stirred at room temperature for 6h. The precipitate formed was collected by filtration then dissolved in aqueous sodium bicarbonate. Acidification to pH3 with 1M HCl led to the formation of a white precipitate which was collected and dried (194mg)

^{1}H NMR (DMSO, δ) 3.2-3.48 (brm, 4H) 3.59 (t, 2H) 3.89 (t, 2H) 6.96 (m, 1H) 7.30 (dd, 1H) 7.85 (m, 1H)

Intermediate 15

6-Chloro-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide

A mixture of racemic 3-amino-5-phenyl-1,3-dihydro-benzo[e][1,4]diazepin-2-one (1g), O-benzotriazol-1-yl-N,N,N',N'-tetramethyluronium hexafluorophosphate (1.51g), triethylamine (0.83ml) and 6-chloro-nicotinic acid (0.63g) in dry DMF (20ml) was stirred at room temperature for 1.5h. Water (200ml) was then added and the mixture stirred vigorously for 10mins. The colourless precipitate was collected by filtration and dried (1.1g)

^{1}H NMR (DMSO, δ) 5.50 (d, 1H) 7.28-7.71 (m, 10H) 8.42 (dd, 1H) 9.01 (d, 1H) 9.99 (d, 1H) 10.95 (s, 1H)

LC/MS RT= 4.96mins, ES+ 391,393

Intermediate 16

Thiomorpholine-1,1-dioxide

9.98 g of thiomorpholine and 14.8 g of triflic anhydride were stirred together in DCM at room temperature for 2 hours. The reaction was then partitioned between 1 M $K_2CO_{3(aq)}$ and DCM. The organic layer was separated and dried by passing through a hydrophobic frit, then concentrated *in vacuo*. 13.82 g of the resultant oil was stirred with 85.2 g of oxone in 50 mL of methanol and 50 mL of water for 18 h at room temperature. The reaction was then filtered and washed with methanol and the filtrate concentrated. This was then partitioned between water and EtOAc and the aqueous layer washed 3 times with EtOAc. The combined organic extracts were then dried ($MgSO_4$) and concentrated to produce a white solid. This was then stirred at room temperature with 40 g of K_2CO_3 in 80 mL of methanol for 18 h. The

methanol was then removed *in vacuo* and the remains partitioned between DCM and sat. $K_2CO_{3(aq)}$. The combined organic extracts were passed through a hydrophobic frit and concentrated *in vacuo* to produce the title compound, 3.51 g.

^{1}H NMR ($CDCl_3$, δ) 1.54 (s, 1H), 2.93-2.97 (m, 4H), 3.24-3.28 (m, 4H).

Intermediate 17

5-{[(2-Methanesulfonyl-ethyl)-methyl-amino]-methyl}-furan-2-carboxylic acid ethyl ester

0.5 g of 5-chloromethyl-furan-2-carboxylic acid ethyl ester and 20 ml of 2 M methylamine solution in THF were stirred at room temperature for 5 days under nitrogen. The solution was then concentrated and purified by SPE. The resultant oil was heated at 200 °C in a microwave with 0.2mL of methanesulfonyl-ethene in 3 mL of acetonitrile for 1 h. The solution was concentrated and purified by chromatography to produce the title compound as a colourless oil.

LC/MS RT = 3.55 min, Found ES^+ = 290

^{1}H NMR ($CDCl_3$, δ) 1.29 (t, 3H), 2.25 (s, 3H), 2.92-2.88 (m, 2H), 2.99 (s, 3H), 3.06-2.99 (t, 2H), 3.6 (s, 2H), 4.26 (q, 2H), 6.28 (d, 1H), 7.04 (d, 1H).

Intermediate 18

5-Dimethylaminomethyl-furan-2-carboxylic acid

0.16ml of a 2 M solution of dimethylamine was added to a stirred suspension of 19.2 mg of sodium hydride in 2 mL of DMF under a nitrogen atmosphere at room temperature for 30 min. Then a solution of 5-chloromethyl-furan-2-carboxylic acid ethyl ester in 2 mL of DMF was added dropwise over a period of 30 min. The reaction was then allowed to stir for 2 days. The solvent was then removed *in vacuo* and 5 mL of EtOH and 0.35ml of 2 M NaOH added and stirred at 80 °C for 40 min. Upon return the reaction was acidified below pH 5.0

and the solvent removed *in vacuo* to produce the title compound to be hydrolysed and then used crude in the next stage

Intermediates 19-23 were prepared in an analogous manner and were used without characterisation in the next synthetic step

Intermediate 19

5-Morpholin-4-ylmethyl-furan-2-carboxylic acid

Intermediate 20

5-(1,1-Dioxo-1λ^6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid

Intermediate 21

5-(4-Methyl-piperazin-1-ylmethyl)-furan-2-carboxylic acid

Intermediate 22

5-(Piperidin-1-ylmethyl)-furan-2-carboxylic acid

Intermediate 23

5-(Pyrrolidin-1-ylmethyl)-furan-2-carboxylic acid

Intermediate 24

3-Cyclopropyl-1,3-dihydro[4,5-b]pyridin-2-one

A mixture of 2-chloro-3-nitro-pyridine (2g), cyclopropylamine (1.13ml) and potassium carbonate (3.48g) in acetonitrile (30ml) was stirred at room temperature for 18h. The mixture was then partitioned between water and ethyl acetate. The dried extracts were evaporated giving a bright yellow solid (2.1g)

This material was then hydrogenated at atmospheric pressure in ethanol (150ml) over palladium on carbon catalyst (10%, 100mg). When hydrogen uptake had ceased the mixture was filtered through celite and evaporated giving a dark gum (1.7g)

This material was then dissolved in dry THF (40ml) and was treated with carbonyl di-imidazole (2.2g) at reflux for 2.5h. The mixture was then partitioned between water and ethyl acetate. The dried organic extract was evaporated leaving a dark gum, which was crystallised from ethyl acetate/petrol giving a colourless solid (1.2g)

^{1}H NMR (DMSO, δ) 0.97-1.04 (m, 4H) 2.92 (m, 1H) 6.97 (dd, 1H) 7.22 (dd,1H) 7.92 (dd, 1H) 10.95 (brs, 1H)

Intermediate 25

2-Morpholin-4-ylmethyl-furan-3-carboxylic acid methyl ester

A mixture of 2-chloromethyl-furan-3-carboxylic acid methyl ester (100mg) and morpholine (0.08ml) in acetonitrile (4ml) was stirred at room temperature for 2h. The mixture was then partitioned between dichloromethane and aqueous sodium bicarbonate solution. The dried organic layer was evaporated giving a yellow oil (75mg)

^{1}H NMR ($CDCl_3$, δ) 2.57 (m, 4H) 3.74 (m, 4H) 3.86 (s, 3H) 3.97 (s, 2H) 6.70 (d, 1H) 7.38 (d, 1H)

Intermediate 26

3-Morpholin-4-ylmethyl-benzoic acid methyl ester

This material was prepared as for Intermediate 25. The product was a colourless oil (210mg)

^{1}H NMR ($CDCl_3$, δ) 2.43 (m, 4H) 3.53 (s, 2H) 3.70 (m, 4H) 3.91 (s, 3H) 7.39 (t, 1H) 7.42 (dd,1H) 7.93 (dt, 1H) 7.99 (brs, 1H)

Intermediate 27

5-Morpholin-4-ylmethyl-isoxazole-3-carboxylic acid methyl ester

5-Methyl-isoxazole-3-carboxylic acid methyl ester (200mg), N-bromosuccinimide (252mg) and bezoyl peroxide (30mg) in dry chloroform (4ml) was stirred and heated to 85C for 5h. The solution was cooled to room temperature and was treated with morpholine (0.27ml). Stirring was continued for 20h and the mixture was then partitioned between water and dichloromethane. The dried organic extract was evaporated and the residue purified on a silica gel SPE cartridge. Elution with dichloromethane followed by dichloromethane:ethanol:0.880 ammonia; 200:8:1 gave a colourless oil (50mg)

^{1}H NMR ($CDCl_3$, δ) 2.46 (m, 4H) 3.64 (m, 4H) 3.67 (s, 2H) 3.90 (s, 3H) 6.55 (s, 1H)

Intermediates 28-30 were prepared in an analogous method to Intermediate 25

Intermediate 28

3-Morpholin-4-ylmethyl-furan-2-carboxylic acid methyl ester

This compound was isolated as a yellow oil (189mg)

^{1}H NMR ($CDCl_3$, δ) 2.45 (m, 4H) 3.65 (m, 4H) 3.71 (s, 2H) 3.85 (s, 3H) 6.56 (d, 1H) 7.45 (d, 1H)

Intermediate 29

3-Morpholin-4-ylmethyl-thiophene-2-carboxylic acid methyl ester

This compound was isolated as yellow oil (197mg).

^{1}H NMR ($CDCl_3$, δ) 2.50 (m, 4H) 3.69 (s, 2H) 3.72 (m, 4H) 3.86 (s, 3H) 6.90 (d, 1H) 7.64 (d, 1H)

Intermediate 30

5-Morpholin-4-ylmethyl-thiophene-2-carboxylic acid methyl ester

This compound was isolated as a yellow oil (214mg).

^{1}H NMR ($CDCl_3$, δ) 2.44 (m, 4H) 3.64 (m, 4H) 3.79 (s, 3H) 3.84 (s, 2H) 7.15 (d, 1H) 7.36 (d, 1H)

Intermediates 25-30 were hydrolysed to the corresponding carboxylic acids before use in the final coupling step of the synthetic sequence

Intermediate 31

4-Fluoro-2-morpholin-4-yl-benzoic acid

2,4-Difluoro-benzoic acid (50mg) and morpholine (0.03ml) in acetonitrile (0.5ml) were heated in a microwave at 200C for 15mins. The solvent was evaporated leaving a dark gum which was used without purification in the next synthetic step.

Intermediate 32

4-Fluoro-2-piperidin-1-yl-benzoic acid

This was prepared in an analogous procedure to Intermediate 31.

Intermediates 33-5 were prepared in an analogous procedure to Intermediate 31 except that 2-fluoro-4-trifluoromethyl-benzoic acid was used.

Intermediate 33

2-Pyrrolidin-1-yl-4-trifluoromethyl-benzoic acid

Intermediate 34

2-Piperidin-1-yl-4-trifluoromethyl-benzoic acid

Intermediate 35

2-Morpholin-4-yl-4-trifluoromethyl-benzoic acid

Intermediates 36 and 37 were prepared in an analogous procedure to Intermediate 31 except that 2-fluoro-5-trifluoromethyl-benzoic acid was used.

Intermediate 36

2-Pyrrolidin-1-yl-5-trifluoromethyl-benzoic acid

Intermediate 37

2-Morpholin-4-yl-5-trifluoromethyl-benzoic acid

Intermediates 38 and 39 were prepared in an analogous procedure to Intermediate 31 except that 4-cyano-2-fluoro-benzoic acid was used.

Intermediate 38

4-Cyano-2-pyrrolidin-1-yl-benzoic acid

Intermediate 39

4-Cyano-2-piperidin-1-yl-benzoic acid

Example 1.

6-(4-Methyl-piperazin-1-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide

Intermediate 15 (50mg) and N-methylpiperazine (0.022ml) in acetonitrile (1ml) containing triethylamine (0.027ml) was heated in a microwave at 200°C for 10mins.The mixture was then partitioned between water and dichloromethane. The dried organic layer was evaporated and the residue purified on a silica gel SPE cartridge. Gradient elution with 5-10% methanol in dichloromethane gave a colourless solid (10mg)

1H NMR (DMSO, d) 2.28 (s, 3H) 2.45 (m, 4H) 3.68 (m, 4H) 5.56 (d, 1H) 6.93 (d, 1H) 7.32-7.72 (m, 10H) 8.20 (dd, 1H) 8.82 (d, 1H) 9.42 (d, 1H) 10.94 (s, 1H)
RT= 3.94mins, ES+ 455

Example 2

3,4,5,6-Tetrahydro-2H-[1,2']bipyridinyl-5'-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 1 except that piperidine was used as the nucleophile. The product was a colourless solid (15mg)

1H NMR (DMSO, d) 1.54-1.63 (brm, 6H) 3.65 (m, 4H) 5.48 (d, 1H) 6.86 (d, 1H) 7.25-7.65 (m, 10H) 8.11 (dd, 1H) 8.75 (d, 1H) 9.32 (d, 1H)
RT= 4.54 mins, ES+ 440

Example 3

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl-benzamide

(S)-3-Amino-5-phenyl-1,3-dihydro-benzo[e][1,4]diazepin-2-one (100mg), O-benzotriazol-1-yl-N,N,N',N'-tetramethyluronium hexafluorophosphate (150mg), 2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid (102mg) and triethylamine (0.083ml) in dry DMF (1ml) was stirred at room temperature for 1h. Water (10ml) was then added and stirring continued for 10mins. The colourless precipitate was collected by filtration and then partitioned between dichloromethane and water. The dried organic phase was evaporated and the residue purified on a silica gel SPE cartridge. Elution with ethyl acetate: petrol 1:1 gave the title compound as a colourless solid (140mg)

^{1}H NMR (DMSO, δ) 3.49 (brs, 8H) 5.48 (d, 1H) 7.31-7.95 (m, 13H) 10.86 (d, 1H) 11.18 (s, 1H)

Example 4

(S)-2-Chloro-4-morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 2-chloro-4-morpholin-4-yl-benzoic acid (86mg) was used. The title compound was a colourless solid (112mg).

^{1}H NMR (DMSO, δ) 3.21 (m, 4H) 3.70 (t, 4H) 5.36 (d, 1H) 6.90-6.97 (m, 2H) 7.21-7.66 (m, 10H) 9.21 (d, 1H) 10.86 (s, 1H)

Example 5

(S)-2-(1,1-Dioxo-4-oxy-1λ6-thiomorpholin-4-yl)-4-fluoro-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl-benzamide

This material was prepared as for Example 3 except that 2-(1,1-dioxo-4-oxy-1λ6-thiomorpholin-4-yl)-benzoic acid (Intermediate 14, 30mg) was used. The title compound was a colourless solid (29mg).

1H NMR (DMSO, d) 3.32-3.98 (m, 8H) 5.34 (d, 1H) 6.99 (dt, 1H) 7.16-7.65 (m, 11H) 9.51 (d,1H) 10.98 (s, 1H)
RT= 5.09mins, ES+ 523

Example 6

(S)-5-Chloro-2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 5-Chloro-2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid (Intermediate 11, 58mg) was used. The title compound was a colourless solid (70mg).

1H NMR (DMSO, d) 3.54 (s, 8H) 5.53 (d, 1H) 7.37-7.75 (m, 11H) 7.90 (d, 1H) 10.84 (d, 1H) 11.24 (s, 1H)
RT= 5.38mins, ES+ 523,525

Example 7

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-5-fluoro-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 5-Fluoro-2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid (Intermediate 12, 54mg) was used. The title compound was a colourless solid (70mg).

1H NMR (DMSO, d) 3.49 (m,8H) 5.47 (d,1H) 7.34-7.69 (m, 12H) 11.12 (d,1H) 11.20 (s, 1H)
RT= 5.19mins, ES+ 507

Example 8

(S)-5-(4-Methyl-piperazin-1-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-(4-Methyl-piperazin-1-ylmethyl)-furan-2-carboxylic acid (Intermediate 21) was used. The title compound was a colourless solid (15mg).

1H NMR (CDCl3, d) 2.23 (s, 3H), 2.43-2.51 (m, 8H), 3.56 (s, 2H), 5.65 (d, 1H), 6.29 (d, 1H), 7.05-7.51 (m, 11H), 7.92 (d, 1H).
RT = 4.10 mins, ES+ 458

Example 9

(S)-5-Pyrrolidin-1-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-(pyrrolidin-1-ylmethyl)-furan-2-carboxylic acid (Intermediate 23) was used. The title compound was a colourless solid (52mg).

1H NMR (CDCl3, d) 1.76-1.77 (m, 4H), 2.60-2.62 (m, 4H), 3.71 (s, 2H), 5.64 (d, 1H), 6.31 (d, 1H), 7.05-7.50 (m, 10H), 7.98 (d, 1H), 8.04 (s, 1H).
RT = 4.09 mins, ES+ 403

Example 10

(S)-5-Piperidin-1-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-(piperidin-1-ylmethyl)-furan-2-carboxylic acid (Intermediate 22) was used. The title compound was a colourless solid (21mg).

1H NMR (CDCl3, d) 1.36-1.45 (m, 2H), 1.53-1.60 (m, 4H), 2.45-2.55 (m, 4H), 3.62 (s, 2H), 5.65 (d, 1H), 6.34 (d, 1H), 7.06-5.52 (m, 10H), 7.81-7.89 (m, 1H), 7.96 (d, 1H).
RT = 4.16 mins, ES+ 443

Example 11

(S)-5-Dimethylaminomethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-dimethylaminomethyl-furan-2-carboxylic acid (Intermediate 18) was used. The title compound was a colourless solid (5mg).

1H NMR (DMSO, d) 2.35 (s, 6H), 3.69 (s, 2H), 5.56 (d, 1H), 6.65 (d, 1H), 7.48-7.85 (m, 10H), 9.1 (d, 1H), 11.13 (s, 1H).
RT = 4.09 mins, ES+ 403

Example 12

(S)-4-Fluoro-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-piperidin-1-yl-benzamide

This material was prepared as for Example 3 except that 4-fluoro-2-piperidin-1-yl-benzoic acid (Intermediate 32) was used. The title compound was a colourless solid (58mg).

1H NMR (DMSO, d) 1.62-1.67 (m, 2H) 1.91-1.99 (m, 4H) 3.08-3.16 (m, 4H) 5.56 (d, 1H) 7.15-7.79 (m, 11H) 8.10-8.13 (m, 1H) 11.08 (s and d, 2H)
RT= 6.02mins, ES+ 457

Example 13

(S)-4-Fluoro-2-morpholino-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 4-fluoro-2-morpholin-4-yl-benzoic acid (Intermediate 31) was used. The title compound was a colourless solid (19mg).

1H NMR (DMSO, d) 2.94-3.00 (m, 4H) 3.71-3.82 (m, 4H) 5.35 (d, 1H) 6.98-7.85 (m, 12H) 10.52 (d, 1H) 10.90 (s, 1H)
RT= 5.34mins, ES+ 459

Example 14

(S)-4-Cyano-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-pyrrolidin-1-yl-benzamide

This material was prepared as for Example 3 except that 4-cyano-2-pyrrolidin-1-yl-benzoic acid (Intermediate 38) was used. The title compound was a colourless solid (13mg).
1H NMR (DMSO, d) 1.87 (brs, 4H) 3.29 (brs, 4H) 5.37(d, 1H) 7.01-7.65 (m, 12H) 9.60 (d, 1H) 10.88 (s, 1H)
RT= 5.45mins, ES+ 450

Example 15

(S)-4-Cyano-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-piperidine-1-yl-benzamide

This material was prepared as for Example 3 except that 4-cyano-2-piperidin-1-yl-benzoic acid (Intermediate 39) was used. The title compound was a colourless solid (27mg).

1H NMR (DMSO, d) 1.32-1.36 (m, 2H) 1.58-1.67 (m, 4H) 2.81-2.89 (m, 4H) 5.25 (d, 1H) 7.10-7.83 (m, 12H) 10.70 (d, 1H) 10.81 (s, 1H)
RT= 5.88mins, ES+ 464

Example 16

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-pyrrolidin-1-yl-4-trifluoromethyl-benzamide

This material was prepared as for Example 3 except that 2-pyrrolidin-1-yl-4-trifluoromethyl-benzoic acid (Intermediate 33) was used. The title compound was a colourless solid (5mg).

1H NMR (DMSO, d) 1.89-1.92 (brs, 4H) 3.29-3.32 (brs, 4H) 5.40 (d, 1H) 6.88 (s, 1H) 6.94 (d, 1H) 7.24-7.67 (m, 10H) 9.56 (d, 1H) 10.89 (s, 1H)
RT= 5.91mins, ES+ 493

Example 17

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-piperidin-1-yl-4-trifluoromethyl-benzamide

This material was prepared as for Example 3 except that 2-piperidin-1-yl-4-trifluoromethyl-benzoic acid (Intermediate 34) was used. The title compound was a colourless solid (14mg).

1H NMR (DMSO, d) 1.53-1.57 (m, 2H) 1.80-1.91 (m, 4H) 3.00-3.14 (m, 4H) 5.46 (d, 1H) 7.30-7.72 (m, 11H) 8.09 (d, 1H) 10.98 (d, 1H) 10.99 (s, 1H)
RT=6.39mins, ES+ 507

Example 18

(S)-2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-4-trifluoromethyl-benzamide

This material was prepared as for Example 3 except that 2-morpholin-4-yl-4-trifluoromethyl-benzoic acid (Intermediate 35) was used. The title compound was a colourless solid (14mg).

1H NMR (DMSO, d) 3.18-3.24 (m, 4H) 3.90-3.96 (m, 4H) 5.52 (d, 1H) 7.36-8.10 (m, 12H) 10.59 (d, 1H) 11.10 (s, 1H)
RT= 5.72mins, ES+ 509

Example 19

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-2-pyrrolidin-1-yl-5-trifluoromethyl-benzamide

This material was prepared as for Example 3 except that 2-pyrrolidin-1-yl-5-trifluoromethyl-benzoic acid (Intermediate 36) was used. The title compound was a colourless solid (8mg).

1H NMR (DMSO, d) 2.00-2.02 (brs, 4H) 3.40-3.43 (brs, 4H) 5.48 (d, 1H) 6.90 (d, 1H) 7.34-7.74 (m, 11H) 9.71 (d, 1H) 10.98 (s, 1H)
RT= 5.84 mins, ES+ 493

Example 20

(S)-2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-5-trifluoromethyl-benzamide

This material was prepared as for Example 3 except that 2-morpholin-4-yl-5-trifluoromethyl-benzoic acid (Intermediate 37) was used. The title compound was a colourless solid (19mg).

1H NMR (DMSO, d) 3.13-3.18 (m, 4H) 3.85-3.90 (m, 4H) 5.46 (d, 1H) 7.30-7.69 (m, 10H) 7.88 (dd, 1H) 8.04 (d, 1H) 10.37 (d, 1H) 11.04 (s, 1H)
RT= 5.72mins, ES+ 509

Example 21

(S)-2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide

This material was prepared as for Example 3 except that 2-morpholin-4-yl-nicotinic acid was used. The title compound was a colourless solid (45mg).

1H NMR (DMSO, d) 3.30-3.36 (m, 4H) 3.82-3.85 (m, 4H) 5.45 (d, 1H) 7.14-7.17 (m, 1H) 7.19-7.71 (m, 9H) 8.07 (dd, 1H) 8.44 (dd, 1H) 10.00 (d, 1H) 11.05 (s, 1H)
RT= 4.86mins, ES+ 442

Example 22

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide

This material was prepared as for Example 3 except that 2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-nicotinic acid (Intermediate 3) was used. The title compound was a colourless solid (10mg).

1H NMR (DMSO, d) 3.25 (t, 2H) 3.40 (t, 2H) 3.75-3.88 (m, 4H) 5.47 (d, 1H) 6.67-6.72 (m, 1H) 7.28-7.67 (m, 8H) 8.24- 8.38 (m, 3H) 9.56 (d, 1H) 10.92 (s, 1H)
RT= 4.43mins, ES+ 508

Example 23

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-3-methyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-3-methyl-benzoic acid (Intermediate 4) was used. The title compound was a colourless solid (65mg).

1H NMR (DMSO, d) 2.36 (s, 3H) 3.24 (brs, 4H) 3.49 (brs, 4H) 5.43 (d, 1H) 7.11-7.68 (m, 12H) 9.61 (d, 1H) 10.99 (s, 1H)
RT= 5.04mins, ES+ 503

Example 24

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-4-methyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-4-methyl-benzoic acid (Intermediate 5) was used. The title compound was a colourless solid (72mg).

1H NMR (DMSO, d) 2.39 (s, 3H) 3.44-3.54 (brm, 8H) 5.46 (d, 1H) 7.14 (d, 1H) 7.31-7.69 (m, 10H) 7.86 (d, 1H) 10.94 (d, 1H) 11.17 (s, 1H)
RT= 5.20mins, ES+ 503

Example 25

(S)-2-(1,1-Dioxo-1λ6-thiomorpholin-4-yl)-6-methyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 2-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-6-methyl-benzoic acid (Intermediate 6) was used. The title compound was a colourless solid (32mg).

1H NMR (DMSO, d) 2.27 (s, 3H) 3.24-3.27 (m, 4H) 3.41-3.43 (m, 4H) 5.56 (d, 1H) 7.03 (d, 1H) 7.11 (d, 1H) 7.25-7.68 (m, 10H) 9.44 (d, 1H) 10.96 (s, 1H)
RT=5.03mins, ES+ 503

Example 26

(S)-2-Chloro-6-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 2-chloro-6-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid (Intermediate 10) was used. The title compound was a colourless solid (51mg).

1H NMR (DMSO, d) 3.43-3.47 (m, 4H) 3.59-3.61 (m, 4H) 5.63 (d, 1H) 7.39-7.83 (m, 12H) 9.86 (d, 1H) 11.14 (s, 1H)
RT= 5.07mins, ES+ 523, 525

Example 27

(S)-3-Cyclopropyl-2-oxo-2,3-dihydro-imidazo[4,5-b]pyridine-1-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

3-Cyclopropyl-1,3-dihydro[4,5-b]pyridin-2-one (Intermediate 24, 35mg), triethylamine (0.028ml) and triphosgene (20mg) were stirred at room temperature in dichloromethane (3ml) for 1h. (S)-3-Amino-5-phenyl-1,3-dihydro-benzo[e][1,4]diazepin-2-one (50mg) was then added, and stirring continued for 18h. The solvent was evaporated and the residue purified on a silica gel SPE cartridge. Elution with dichloromethane:ethanol:0.880 ammonia; 200:8:1 gave a colourless solid (3mg)

1H NMR (DMSO, d) 0.88-1.09 (m, 4H) 2.92 (m ,1H) 5.25 (d, 1H) 7.06-7.71 (m, 10H) 8.08 (m, 2H) 9.94 (d,1H) 11.08(s,1H)
RT= 4.90mins, ES+ 453

Example 28

(S)-3-(4-Methyl-piperazine-1-sulfonyl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 3-(4-methyl-piperazine-1-sulfonyl)-benzoic acid (Intermediate 7) was used. The title compound was a pale yellow solid (23mg).

1H NMR (CDCl3, d) 2.19 (s, 3H), 2.39-2.43 (m, 4H), 2.95-3.05 (m, 4H), 5.68 (d, 1H), 6.5 (s, 1H), 7.13 (t, 2H), 7.19 (s, 1H), 7.32-7.83 (m, 8H), 8.08-8.11 (m, 2H), 8.28-8.29 (m, 1H).
RT = 4.25 mins, ES+ 518

Example 29

(S)-4-(4-Methyl-piperazin-1-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 4-(4-methyl-piperazine-1-yl)-benzoic acid was used. The title compound was a colourless solid (46mg).

1H NMR (CDCl3, d) 2.30 (s, 3H), 2.50-2.54 (m, 4H), 3.26-3.30 (m, 4H), 5.70 (d, 1H), 6.86 (d, 2H), 7.14 (t, 1H), 7.17-7.50 (m, 8H), 7.74 (d, 1H), 7.80 (d, 2H), 8.25-8.40 (m, 1H).
RT = 4.16 mins, ES+ 454

Example 30

(S)-N-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-3-(piperidine-1-sulfonyl)-benzamide

This material was prepared as for Example 3 except that 3-piperidine-1-sulfonyl-benzoic acid (Intermediate 8) was used. The title compound was a colourless solid (35mg).

1H NMR (CDCl3, d) 1.35-1.38 (m, 2H), 1.57-1.65 (m, 4H), 2.91-2.99 (m, 4H), 5.70 (d, 1H), 7.14 (t, 2H), 7.19 (s, 2H), 7.31-7.84 (m, 7H), 8.04-8.12 (m, 2H), 8.28-8.29 (m, 1H), 8.41 (s, 1H).
RT = 5.47 mins, ES+ 503

Example 31

(S)-3-(Morpholine-4-sulfonyl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 3-(morpholine-4-sulfonyl)-benzoic acid (Intermediate 9) was used. The title compound was a colourless solid (29mg).

1H NMR (CDCl3, d) 2.97-3.00 (m, 4H), 3.66-3.70 (m, 4H), 5.68 (d, 1H), 7.10-8.18 (m, 13H), 8.29-8.31 (m, 2H).

RT = 5.06 mins, ES+ 505

Example 32

(S)-5-Morpholin-4-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-morpholin-4-ylmethyl-furan-2-carboxylic acid (Intermediate 19) was used. The title compound was a colourless solid (35mg).

1H NMR (CDCl3, d) 2.46-2.49 (m, 4H), 3.55 (s, 2H), 3.66-3.70 (m, 4H), 5.65 (d, 1H), 6.30 (d, 1H), 7.06-7.51 (m, 10H), 7.95 (d, 1H), 8.38 (s, 1H).
RT = 4.28 mins, ES+ 445

Example 33

(S)-5-Hydroxymethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that the hydrolysis product of 5-chloromethyl-furan-2-carboxlic acid ethyl ester was used. The title compound was a colourless solid (48mg).

1H NMR (CDCl3, d) 2.78 (s, 1H), 4.55-4.56 (m, 2H), 5.63 (d, 1H), 6.25 (d, 1H), 7.00 (d, 1H), 7.09 (t, 2H), 7.15-7.49 (m, 7H), 8.10 (d, 1H), 8.46 (s, 1H).
RT = 4.54 mins, ES+ 376

Example 34

(S)-5-(1,1-Dioxo-1λ6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-(1,1-Dioxo-1λ6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid (Intermediate 20) was used. The title compound was a colourless solid (192mg).

1H NMR (CDCl3, d) 3.00-3.10 (m, 8H), 3.68 (s, 2H), 5.65 (d, 1H), 6.32 (d, 1H), 7.06-7.50 (m, 10H), 7.95 (d, 1H), 8.08-8.16 (s, 1H).
RT = 4.65 mins, ES+ 493

Example 35

(S)-2-Chloro-4-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 2-chloro-4-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid (Intermediate 1) was used. The title compound was a colourless solid (41mg).

1H NMR (DMSO, d) 3.15 (brs, 4H) 3.92 (brs, 4H) 5.41 (d, 1H) 7.10-7.68 (m, 12H) 9.26 (d, 1H) 10.92 (s, 1H)
RT= 4.70mins, ES+ 523, 525

Example 36

(S)-2-Chloro-5-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 2-chloro-5-(1,1-dioxo-1λ6-thiomorpholin-4-yl)-benzoic acid (Intermediate 2) was used. The title compound was a colourless solid (69mg).

1H NMR (DMSO, d) 3.14 (brs, 4H) 3.81 (brs, 4H) 5.37 (d, 1H) 7.08-7.63 (m, 12H) 9.56 (d, 1H) 10.84 (s, 1H)
RT= 4.76mins, ES+ 523,525

Example 37

(S)-5-{[(2-Methanesulfonyl-ethyl)-methyl-amino]-methyl}-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl-amide

This material was prepared as for Example 3 except that 5-{[(2-methanesulfonyl-ethyl)-methyl-amino]-methyl}-furan-2-carboxylic acid ethyl ester (Intermediate 17) was used. The title compound was a colourless solid (87mg).

1H NMR (DMSO, d) 2.05 (s, 3H), 2.61 (t, 2H), 2.84 (s, 3H), 3.12 (t, 2H), 3.48 (s, 2H), 5.21 (d, 1H), 6.34 (d, 1H), 7.05-7.39 (m, 9H), 7.50 (td, 1H), 8.77 (d, 1H), 10.78 (s, 1H).
RT = 4.78 mins, ES+ 495

Example 38

(S)-2-Pyridin-3-yl-thiazole-4-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 2-pyridin-3-yl-thiazole-4-carboxylic acid was used. The title compound was a colourless solid (55mg).

1H NMR (DMSO, d) 5.64 (d, 1H) 7.48-7.86 (m, 10H) 8.66 (dt, 1H) 8.73 (s, 1H) 8.93 (dd,1H) 9.31 (d, 1H) 9.47 (d, 1H) 11.28 (s, 1H)

RT=4.70mins, ES+ 440

Example 39

(S)-2-Pyridin-4-yl-thiazole-4-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 2-pyridin-4-yl-thiazole-4-carboxylic acid was used. The title compound was a colourless solid (54mg).

1H NMR (DMSO, d) 5.36 (d, 1H) 7.19-7.58 (m, 9H) 7.96 (dd, 2H) 8.53 (s, 1H) 8.69 (dd, 2H) 9.02 (d, 1H) 11.01 (s, 1H)
RT= 4.69mins, ES+ 440

Example 40

(S)-4-Methyl-2-pyrazin-2-yl-thiazole-5-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 4-methyl-2-pyrazin-2-yl-thiazole-5-carboxylic acid was used. The title compound was a colourless solid (67mg).

1H NMR (DMSO, d) 2.56 (s, 3H) 5.25 (d, 1H) 7.10-7.49 (m, 9H) 8.58-8.63 (s+dd, 2H) 9.16 (d, 1H) 9.38 (d, 1H) 10.78 (s, 1H)
RT= 4.82mins, ES+ 455

Example 41

(S)-2-Morpholin-4-ylmethyl-furan-3-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 2-morpholin-4-ylmethyl-furan-3-carboxylic acid (Intermediate 25) was used. The title compound was a colourless solid (24mg).

1H NMR (DMSO, d) 2.58 (brm, 4H) 3.67 (brm, 4H) 3.91 (s, 2H) 5.45 (d, 1H) 6.88 (d,1H) 7.33-7.75 (m, 10H) 10.95 (s, 1H) 11.01 (d, 1H)
RT= 5.04mins, ES+ 445

Example 42

(S)-3-Morpholin-4-ylmethyl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Example 3 except that 3-morpholin-4-ylmethyl-benzoic acid (Intermediate 26) was used. The title compound was a colourless solid (24mg).

1H NMR (DMSO, d) 2.39 (brm, 4H) 3.55 (s, 2H) 3.60 (brm, 4H) 5.51 (d, 1H) 7.28-7.71(m, 11H) 7.93 (s, 1H) 7.97 (s, 1H) 9.50 (d, 1H) 10.93 (s, 1H)
RT= 4.86mins, ES+ 455

Example 43

(S)-5-Morpholin-4-ylmethyl-isoxazole-3-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-morpholin-4-ylmethyl-isoxazole-3-carboxylic acid (Intermediate 27) was used. The title compound was a colourless solid (11mg).

1H NMR (DMSO, d) 2.93 (m, 4H) 3.46 (m, 4H) 3.66 (brs, 2H) 5.26 (d, 1H) 6.77 (s, 1H) 7.13-7.38 (m, 9H) 9.17 (d, 1H) 10.90 (s, 1H)

RT= 4.75mins, ES+ 446

Example 44

(S)-3-Morpholin-4-ylmethyl-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 3-morpholin-4-ylmethyl-furan-2-carboxylic acid (Intermediate 28) was used. The title compound was a colourless solid (20mg).

1H NMR (DMSO, d) 2.52 (brm, 4H) 3.62 (brs, 4H) 3.67 (m, 2H) 5.39 (d, 1H) 6.67 (d, 1H) 7.25-7.71 (m, 9H) 7.84 (d, 1H) 10.93 (s, 1H) 11.34 (d, 1H)
RT= 4.96mins, ES+ 445

Example 45

(S)-5-Pyridin-2-yl-thiophene-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-pyridin-2-yl-thiophene-2-carboxylic acid was used. The title compound was a colourless solid (32mg).

1H NMR (DMSO, d) 5.58 (d, 1H) 7.37-7.77 (m, 10H) 7.96-7.99 (m, 2H) 8.10 (d, 1H) 8.32 (d, 1H) 8.67 (d, 1H) 9.81 (d, 1H) 11.03 (s, 1H)
RT= 4.91mins, ES+ 439

Example 46

(S)-2-Methyl-4-(morpholin-4-sulfonyl)-furan-3-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 2-methyl-4-(morpholin-4-sulfonyl)-furan-3-carboxylic acid was used. The title compound was a colourless solid (75mg).

1H NMR (DMSO, d) 2.77 (s, 3H) 3.26 (m, 4H) 3.85 (m, 4H) 5.60 (d, 1H) 7.43-7.83 (m, 9H) 8.23 (s, 1H) 9.68 (d, 1H) 11.07 (s, 1H)
RT= 4.90mins, ES+ 509

Example 47

(S)-6-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-nicotinamide

This material was prepared as for Example 3 except that 6-morpholin-4-nicotinic acid was used. The title compound was a colourless solid (28mg).

1H NMR (DMSO, d) 3.58-3.61 (m, 4H) 3.70-3.73 (m, 4H) 5.51 (d, 1H) 6.89 (d, 1H) 7.24-7.71 (m, 9H) 8.19 (dd, 1H) 8.80 (d, 1H) 9.39 (d, 1H) 10.89 (s, 1H)
RT= 4.59mins, ES+ 442

Example 48

(S)-3-Morpholin-4-ylmethyl-thiophene-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 3-morpholin-4-ylmethyl-thiophene-2-carboxylic acid (Intermediate 29) was used. The title compound was a colourless solid (34mg).

1H NMR (DMSO, d) 2.43 (m, 4H) 3.59 (m, 4H) 3.70 (s, 2H) 5.45 (d, 1H) 7.05 (d, 1H) 7.24-7.70 (m, 9H) 8.05 (d, 1H) 9.54 (d, 1H) 10.92 (s, 1H)

RT= 5.02mins, ES+ 461

Example 49

(S)-5-Morpholin-4-ylmethyl-thiophene-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide

This material was prepared as for Example 3 except that 5-morpholin-4-ylmethyl-thiophene-2-carboxylic acid (Intermediate 30) was used. The title compound was a colourless solid (41mg).

1H NMR (DMSO, d) 2.28 (brm, 4H) 3.38 (brm, 4H) 3.56 (s, 2H) 5.16 (d, 1H) 6.90 (d, 1H) 7.04-7.44 (m, 9H) 7.52 (d, 1H) 10.68 (s, 1H) 11.82 (d, 1H)
RT= 5.33mins, ES+ 461

Example 50

2-Morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

This material was prepared as for Intermediate 15 except that 2-morpholin-4-yl-benzoic acid (49mg) was used. The product was a colourless solid (33mg)

1H NMR (DMSO, d) 3.01-3.12 (m, 4H) 3.86-3.93 (m, 4H) 5.44 (d, 1H) 7.21-7.71 (m, 12H) 7.93 (dd, 1H) 10.99 (d, 1H) 11.02 (s, 1H)
RT=5.47, ES+441

Example 51

(S)- 5-Phenyl-oxazole-4-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide

(S)-3-Amino-5-phenyl-1,3-dihydro-benzo[e][1,4]diazepin-2-one (60mg), triethylamine (0.037ml) and 5-phenyl-oxazole-4-carbonyl chloride (50mg) in THF (3ml) were stirred at room temperature for 2h. The mixture was then partitioned between water and dichloromethane. The dried organic phase was evaporated and the residue purified on a silica gel SPE cartridge. Elution with dichloromethane:ethanol:0.880 ammonia; 400:8:1 gave the title compound as a colourless solid (42mg).

^{1}H NMR (DMSO, δ) 5.40 (d, 1H) 7.27-7.70 (m, 12H) 8.22-8.26 (m, 2H) 8.72 (s, 1H) 8.88 (d, 1H) 11.14 (s, 1H)
RT=5.22, ES+423.49

Example 52

1-(2-Oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-3-(4-phenoxy-phenyl)-urea

Racemic 3-Amino-5-phenyl-1,3-dihydro-benzo[e][1,4]diazepin-2-one (30mg) and 1-isocyanato-4-phenoxy-benzene (0.022ml) in dry THF (4ml) was stirred at room temperature for 18h. The mixture was then partitioned between water and dichloromethane. The dried organic layer was evaporated and the residue triturated from dichloromethane/diethyl ether giving the title compound as a white solid (25mg)

1H NMR (DMSO, d) 5.23 (d, 1H) 6.98-7.03 (m ,3H) 7.11 (t, 1H) 7.33-7.58 (m ,13H) 7.71 (dt, 1H) 9.18 (s, 1H) 11.03 (brs, 1H)
RT=5.57, ES+463.45

Example 53

3-[1-(3-Methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-2-oxo-2,3-dihydro-benzoimidazol-1-sulfonic acid dimethylamide, 1-Methanesulfonyl-3-[1-(3-methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one, 3-[1-(3-Methyl-butyl)-1H-benzoimidazol-2-

ylmethyl]-2-oxo-2,3-dihydro-benzoimidazol-1-carboxylic acid benzylamide, 5-{3-[1-(3-Methanesulfonyl-propyl)-1H-benzoimidazol-2-ylmethyl]-2-oxo-2,3-dihydro-benzoimidazol-1-yl}-pentanenitrile, 7-[2-(3-Isopropenyl-2-oxo-2,3-dihydrobenzoimidazol-1-ylmethyl)-benzoimidazol-1-yl]-heptanenitril, 1-Ethyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one, 1-Ethyl-3-[1-(2-hydroxy-2-phenyl-ethyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one, 1-Isopropenyl-3-[1-(3-oxo-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one, 1-(4-Hydroxy-benzyl)-3-[1-(3-methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one, 1-Isopropenyl-3-[1-(3-methyl-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-benzoimidazol-2-one, 1-Cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one and 1-Isopropenyl-3-(1-propyl-1H-benzoimidazol-2-ylmethyl)-1,3-dihydro-imidazo[4,5-c]pyridine-2-one are prepared as described in WO00195910

Example 54

{2-[2-(1,2-Dihydro-benzotriazol-1-ylmethyl)-benzoimidazol-1-yl]]ethyl}-diethyl-amine is prepared as described in WO00004900.

Example 55

{2-[2-(3-Iodo-2,3-dihydro-indazol-1-ylmethyl)-benzimidazol-1-yl]-ethyl}-dimethyl-amine is prepared as described in WO03053344.

Example 56

Bis(5-amidino-2-benzimidazolyl)-methane is prepared as described in US4,324794.

Example 57

2-{2-[1-[1-(2-Amino-ethyl)-piperidin-4-ylamino]-4-methyl-benzoimidazol-1-ylmethyl}-6-methyl-pyridin-3-ol is prepared as described in WO0100612.

Activity Example 1

Determination of RSV fusion inhibitor activity

RSV enters the host cell via attachment to and fusion with the host cell membrane. The effect of an inhibitor on the specific virus-cell fusion event can be qualitatively determined by using a fluorescence de-quenching system.

The design of this assay takes advantage of the fact that RSV binds to cells at 4°C and at 37°C but that fusion may only occur at concentrations above 18°C.

RSV labelled with octadecyl rhodamine dye (R18) is pre-incubated with Hep-2 cells seeded in a 6-well plate for 1 hour at 4°C to allow binding to occur. Unattached virus is removed by washing the cell monolayer. The inhibitor is then added to the virus-cell complexes prior to transferring the plates to 37°C for 1 hour in order to induce fusion.

Virus-cell fusion can be observed directly under a fluorescence microscope. Fluorescence emission is quenched when 2 identical fluorophores are in close proximity. Upon fusion of the labelled virus with the cell membrane, the distance between fluorophores is increased due to dye spread and there is a decrease in quenching. This is observed as an increase in fluorescence intensity of R18. It therefore follows that inhibition of fusion would lead to a decrease in fluorescence of R18 compared to untreated control. Where the fluorescent yield of R18 in the presence of inhibitor is comparable to the untreated control this would suggest the inhibitor were not exerting its effects on the fusion protein.

Activity Example 2

Determination of RSV replication inhibitor activity

The inner 60 wells of 96 well tissue culture plates are seeded with Hep-2 cells at $4x10^4$ cells/well for compound activity and toxicity studies in 100µl of medium and incubated at 37°C overnight or until nearing confluency.

Cells are infected with 25 µl RSV, e.g. the RSS strain, previously titrated to give 80% cell kill. To each well 25µM of test compound are added. The final DMSO concentration is 0.5%. Some 200 µl of sterile distilled water is added to the outer wells of the plate and incubated at 37°C for 6 days. Some 0.25 µl/ml PMS are added to stock XTT solution, final conc. 25 µM PMS. Then 25 µl warmed XTT/PMS solution is added to each well and incubated for 1 hour at 37°C.

Maximum OD_{450nm} reading (uninfected, untreated control cells) corresponds to 100% inhibition. Minimum OD_{450nm} readings (infected control cells) corresponds to 0% inhibition. Log10 concentration is plotted against OD_{450nm} and IC_{50} values are calculated from either reading 50% value from graph or using regression analysis.

Activity Example 3

Synergistic action between RSV fusion inhibitor and anti-RSV benzodiazepines

ELISA experiments were carried out on the combined effect of potent benzodiazepine RSV replication inhibitor 2-chloro-4-morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide or 5-(1,1-dioxo-1λ6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-IH-benzo[e][1,4]diazepin-3-yl)-amide (compound A) with one RSV fusion inhibitor selected from 1-cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one (compound

B) or 1-isopropenyl-3-(1-propyl-1H-benzoimidazol-2-ylmethyl)-1,3-dihydro-imidazo[4,5-c]pyridine-2-one (compound B)

ELISA PROTOCOL

Mouse monoclonal antibodies to the phosphoprotein (P), nucleocapsid (N) & fusion (F) proteins of RSV and a rabbit anti-mouse- horseradwash peroxidase (HRP) conjugated secondary antibody were used to demonstrate a reduction in RSV antigen via conversion of the o-phenylene diamine dihydrochloride (OPD) substrate to a coloured product. This was quantified by optical density (OD) measurement.

Method

This assay was set up using all 96 wells of flat-bottomed 96-well plates. The outer wells were not subjected to any greater amount of evaporation than the inner wells during the 3 day assay period. (ie. No "edge effect" seen).

Plates were set up one day before addition of virus and compounds. The assay then ran for 3 days with ELISA development taking place on the 4th day.

Day 0

Set up of Assay Plates

All 96 wells of a microtitre plate were seeded at a density of $5x10^3$ Hep-2 cells/well in 100µl/well of Growth Medium (GM) consisting of Dulbecco's MEM (DMEM) with Glutamax-1, Sodium Pyruvate, 1000 mg/l glucose and pyridoxine (Invitrogen, catalogue number 21885-025) and supplemented with 10%FBS. (See Plate 1).

In tissue culture, the cells adhere to the tissue culture flask and were grown at 37°C, 5% CO_2 until 90% confluent.

Monolayers were washed with 20ml sterile PBS to remove serum and treated with 1ml trypsin to detach cells from the flask.

Cells were suspended in a small known volume of growth media and counted using a haemocytometer. The cell suspension was made up to the desired concentration in growth

medium and added to wells by multichannel pipette. Brief, gentle shaking encouraged the cells to disperse more evenly across the well.

Plate 1

cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells
cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells
cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells
cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells
cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells
cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells
cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells
cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells	cells

Plates were kept undisturbed at 37°C in a 5% CO_2 atmosphere for 24hrs during which time the cells settle to form an even cell monolayer.

Day 1

Addition of Virus

A frozen vial of RSV (RSS strain provided by Virogen Ltd) stock solution was removed from the -80 freezer or liquid nitrogen store and diluted to a known Multiplicity of Infection (m.o.i) in Growth Medium.

The m.o.i. was calculated by prior titration of the virus stock (by the ELISA assay method) as the virus input required to achieve a window of at least 0.8 OD units between infected and uninfected control wells.

$$\textbf{Multiplicity of Infection} = \frac{\textbf{plaque forming units per well (pfu/well)}}{\textbf{number of cells per well}}$$

50μl of diluted virus was added to infected wells by multichannel pipette; 50μl of Growth Medium was added to uninfected, cell control wells by multichannel pipette.

Sides of plates were marked with stripes to identify plates in the event of lids becoming separated.

Plates were incubated at 37°C for 1hr to allow virus adsorption.

Compound Dilutions

Compound "A" was titrated horizontally across the plate and Compound "B" was titrated vertically down the plate, creating a chequerboard. The 2 compounds were titrated at either ½-log or doubling dilutions either across (horizontally) or down the plate (vertically) in the presence of virus. Each compound dilution was set up in duplicates or triplicates. For triplicates 3 identical plates were set up. Duplicates were set up as dublicate wells on the same plate. The dilution range covered concentrations from just above the compound IC50 to below the compound IC50 and included a 0μM control for each compound.
Compounds were made up in a separate microtitre plate at 8x strength in GM containing 2% DMSO (a final DMSO concentration in the assay of 0.5%). 25μl of the Compound "A" dilution series and 25μl of the Compound "B" dilution series were then transferred to the appropriate wells of the assay plate by multichannel pipette, according to the marked out chequerboard.
25μl of GM (containing 2% DMSO) was added to wells receiving 0μM Compound "A" or 0μM Compound "B". 50μl GM (containing 2% DMSO) was added to wells containing neither compound.

Virus infected, untreated wells served as the virus control (VC); Uninfected, untreated wells serve as the cell control (CC). The difference in absorbance between CC and VC wells constitutes the assay window.

Plates were incubated at 37°C, 5% CO_2 for 3 days.

ELISA Stage

Day 4

Media was tapped out from wells directly into Virkon (1% solution in water) and plates were washed by immersing in a plastic box containing PBS.

50µl/well of 75%/25% vol/vol acetone/methanol fixative was added by multichannel pipette and left for 3mins.
Acetone/methanol was discarded from wells into Virkon and wells were washed with PBS as above.

Some 200µl of blocking solution (2% Marvel in PBS containing 0.05% Tween) was added per well by multichannel pipette. Plates were incubated at 37°C in a shaking incubator for 60mins.

Block solution was discarded down the sink and diluted primary antibody was added directly to wells (ie. no washing required).
RSV mouse monoclonal antibody NCL-RSV3 (Novocastra) was diluted 1/400 in PBS/2% Marvel/0.05% Tween and 50µl was added per well. Plates were incubated at 37°C in a shaking incubator for 90mins.

Antibody was discarded down the sink and plates were washed 4 times by immersion in PBS/0.05% Tween.
DAKo rabbit anti-mouse HRP conjugate (DAKO catalogue number P0260) was diluted 1/1000 in PBS/2% Marvel/0.05% Tween and 50µl was added per well. Plates were incubated at 37°C in a shaking incubator for 60mins.

Antibody was discarded down the sink and plates were washed 6 times by immersion in PBS/0.05% Tween.

Substrate (SigmaFast OPD) was prepared in advance by dissolving 1 urea tablet in 20mL water. 1 OPD tablet was added to the urea solution just prior to use (NB. OPD was light sensitive) and vortexed to mix. 50µl of substrate was added per well.

The reaction was stopped by addition of 25µl/well of 20% sulphuric acid, once sufficient colour had developed but while cell control background was still low (~5 minutes).

Plates were read on a SpectraMax (Molecular Devices) spectrophotometer at wavelength 490nm and utilize the SOFTmax Pro software package.

The wells were emptied, washed in tap water and the monolayers stained with 50µl/well of 2% crystal violet in 20% methanol/water for at least 1 hour. The wells were then washed and air-dried and the monolayers examined under the microscope for indications of cell toxicity.

Results

SOFTmax data files were exported to Excel. Data handling used Excel templates written in-house for plotting dose response curves graphically and calculating IC50 values from the curves obtained.

All replicate wells were meaned. The assay window was calculated by subtracting the meaned cell control (CC) from the meaned virus control (VC). For each compound, the meaned CC was subtracted from the meaned values for each concentration point. The % of control was then calculated for each concentration point as a percentage of the window. % of control was plotted against compound concentration. A straight line was fitted to the curve and the slope and intercept functions were used to calculate the IC50.

The IC50 for Compound "A" was calculated for each background concentration of Compound "B". Similarly, the IC50 for Compound "B" was calculated for each background concentration of Compound "A".

Example 3a

2-Chloro-4-morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide (Compound A) in combination with 1-cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one (Compound B)

Compound A has an ELISA IC50 of 1.6µM against the RSV RSS strain.
Compound B has an ELISA IC50 of 0.015µM against the RSV RSS strain.

In combination, at concentrations of Compound A below its IC50 the IC50 of Compound B is reduced from 0.15μM to at least 0.003μM (5 -fold decrease). At concentrations of Compound B below its IC50 the IC50 of Compound A is reduced from 1.6μM to at least 1μM (1.6-fold decrease).

Example 3b

5-(1,1-Dioxo-1λ6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide (Compound A) in combination with 1-cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one (Compound B)

Compound A has an ELISA IC50 of 3.5μM against the RSV RSS strain.
Compound B has an ELISA IC50 of 0.06μM against the RSV RSS strain.

In combination, at concentrations of Compound A below its IC50, the IC50 of Compound B is reduced from 0.06μM to at least 0.006μM (10-fold decrease). At concentrations of Compound B below its IC50 the IC50 of compound A is reduced from 3.5μM to at least 0.312μM (11.2-fold decrease).

The formula below can be used to identify a synergistic interaction.

FIC = Fractional Inhibitory concentration
Compares the activity of a compound in combination (Compound A + Compound B) with the activity of the compound alone (Compound A or Compound B).

$$\mathbf{FIC} = \frac{\textbf{Lowest IC50 Cpd A}^{\textbf{COMBINATION}}}{\textbf{IC50 Cpd A}^{\textbf{ALONE}}} + \frac{\textbf{Lowest IC50 Cpd B}^{\textbf{COMBINATION}}}{\textbf{IC50 Cpd B}^{\textbf{ALONE}}}$$

where FIC value < 0.5 SYNERGY

0.5 – 1.0	ADDITION
1.0 – 2.0	INDIFFERENCE
>2.0	ANTAGONISM

FIC for 2-Chloro-4-morpholin-4-yl-N-(2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-benzamide in combination with 1-cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one: 0.3

FIC for 5-(1,1-Dioxo-1λ6-thiomorpholin-4-ylmethyl)-furan-2-carboxylic acid (2-oxo-5-phenyl-2,3-dihydro-1H-benzo[e][1,4]diazepin-3-yl)-amide (Compound A) in combination with 1-cyclopropyl-3-[1-(4-hydroxy-butyl)-1H-benzoimidazol-2-ylmethyl]-1,3-dihydro-imidazo[4,5-c]pyridin-2-one : 0.14